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# THE NEPHROGRAPHIC EFFECT IN DEPRESSED TUBULAR EXCRETION OF UMBRADIL

by

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It is known from clearance studies that if two compounds, both of which are excreted by the tubules, are presented to the tubules simultaneously one may depress the excretion of the other. This is supposed to depend on the fact that tubular excretion, unlike glomerular filtration, is an active cellular process with restricted capacity and, unlike tubular resorption, has only one transport system for different substances. The occupation of the tubules with one substance therefore depresses the excretion of other similarly excreted substances presumably as a result of competition within the transport system rather than an inhibitory or toxic action (SMITH 1951).

Substances excreted by the tubules and which depress the excretion of each other are *e. g.* our common iodine compounds (Diodrast, Umbradil), PAH (paraminohippuric acid), phenol red (phenolsulphonephtalein), and penicillin.

JOSEPHSON & KALLAS (1953) have examined the effect of PAH on the iodine concentration in the kidney tissue when PAH was injected before the contrast medium at urographies in rabbits. They found a reduction of the iodine concentration to one half or a third compared with control animals. This agrees with clearance determinations.

Knowing these facts, the present authors have by urography in rabbits investigated the nephrographic effect when the contrast medium excretion has been depressed after intravenous injection of phenol red (10 % solution), or PAH (20 % solution of sodium paraminohippuric acid). The intention has been to study to what degree the nephrographic

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effect may be influenced. In order to obtain as high a nephrographic effect as possible the intrapelvic pressure in the left kidney of the rabbits was increased.

The examinations have been performed as described in detail in a previous paper by EDLING, HELANDER & RENCK 1954. The rabbits were anaesthetized with ether through a catheter inserted into the trachea. A cannula was inserted into the carotid artery on one side for kymographic registration of the blood pressure. For increasing the intrapelvic pressure a polyethylene catheter was introduced distally in the left ureter in a cranial direction; when an increase in the pressure was required the catheter was elevated vertically until a hydrostatic balance between the glomerular filtration pressure and the intrapelvic pressure was obtained, indicating that the filtration process had ceased.

The injections of phenol red, PAH, and Umbradil were made into a vein of an ear. Films of the kidneys were obtained at intervals of some minutes.

Thirteen healthy rabbits were examined; in 6, Umbradil alone was injected, in the other animals phenol red (one animal) or PAH preceded the Umbradil.

In the first series three animals were injected with 8 ml Umbradil 50 %, two of them twice with a half-hour interval; the other three received 3 to 4 ml Umbradil 35 %, and in addition one of them received 1 ml Umbradil 35 % half an hour before the second dose. An intense nephrographic effect occurred in the left kidney (Fig. 1) after all injections except one. There was no detectable difference between the three animals injected with the higher dose and those injected with the lower. The single poor nephrographic effect was produced by the injection of only 1 ml Umbradil 35 %; in this same animal when 3 ml were injected the nephrographic effect was marked. When contrast filled, the density of the pelves was of a high degree (Fig. 1). At the end of the examination with the counter-pressure released and the blood pressure decreased to less than 70 mm Hg, a nephrographic effect was visible in both kidneys of all animals (EDLING, HELANDER & RENCK 1954).

In the second series six rabbits were injected with 5 to 9 ml PAH and one with 5 ml phenol red 5 to 12 minutes before the contrast medium. Six of the animals were then injected with 5 to 8 ml Umbradil 35 %, one of them twice at 25 min. intervals, and the seventh with 3 ml Umbradil 35 %. In all cases, only a slight nephrographic effect appeared in the left kidney with very slight differences between the various animals (Fig. 2 and Fig. 3, top view). If the left pelvis was contrast-filled during the counter-pressure its density was also low. At the end of the examination after the counter-pressure was released and the blood pressure decreased to less than 70 mm Hg, a nephrographic effect of moderate degree

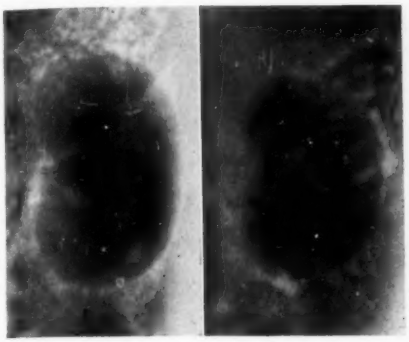

Fig. 1. Left kidneys of two rabbits during urography with catheter and counter-pressure in the ureter. Injection of 3 ml Umbradil 35 %. Marked nephrographic effect. Contrast-filled pelvis of high density in left view.

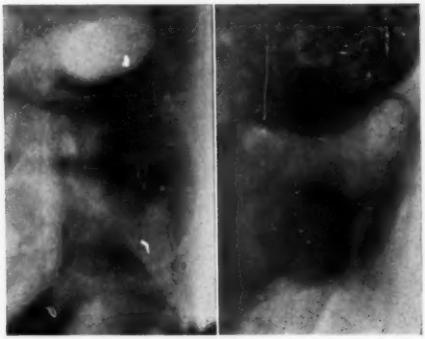

Fig. 2. Left kidneys of two rabbits during urography with catheter and counter-pressure in the ureter. Injection of 5 ml PAH before 5 ml Umbradil 35 %. Only slight nephrographic effect compared with fig. 1.

in both kidneys appeared in one animal (Fig. 3, bottom view) of low degree in one, and absent in five animals. The intervals in time between the injections of phenol red or PAH, and the lowering of the blood pressure were 50, 43, and 20 to 30 minutes, respectively, in the three groups.

The comparison between the two series demonstrates a distinct difference in the degree of nephrographic effect. The injection of the contrast medium alone produces a much greater accumulation of the contrast material in the obstructed kidney than when preceded by phenol red, or PAH. This agrees with the determinations of clearance of substances presented to the tubules simultaneously and with the results of JOSEPHSON & KALLAS 1953. According to these authors, however, 5 ml PAH was too small an amount to have a depressive effect on the renal accumulation. In the present study there was no marked difference in the depressive effect of PAH doses varying from 5 to 9 ml. The difference in the results may be explained by variation in technique as JOSEPHSON & KALLAS did not hinder the excretion of the PAH and the contrast medium.

The theory that the nephrographic effect is to a great part dependent upon the function of the tubular cells and the accumulation of the contrast medium in them is supported by the following. At urography, all of the contrast medium passing the kidneys in vessels, cells, and ducts, contributes to an increase in the radiographic density of the renal tissue on which also the nephrographic effect depends. In the present series

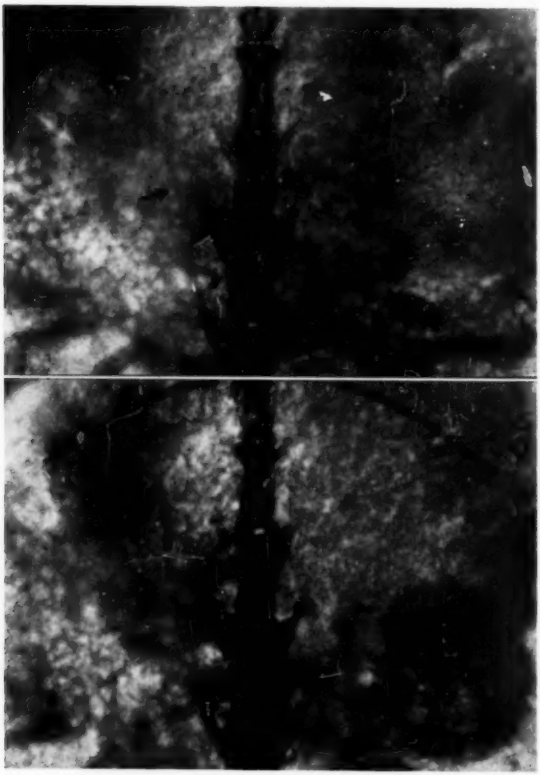

Fig. 3. Urography of rabbit. *Top:* 19 and 14 min. after injection of 5 ml PAH and 5 ml Umbradil 35 % respectively. Catheter and counter-pressure in left ureter. No nephrographic effect visible in left kidney; contrast urine of low density in right ureter. *Bottom:* 54 min. after PAH injection and 26 min. after release of counter-pressure and injection of further 6 ml Umbradil 35 %. Blood pressure fall to 45 mm Hg. Intense nephrographic effect in both kidneys. Contrast urine of higher concentration in right ureter.

the intravascular content of the iodine compound may be discounted due to its low plasma concentration. There remains the contrast medium in the tubular cells (ENGSTRÖM & JOSEPHSON 1953), and in the tubular lumina.

In the first series the tubular transport of the contrast medium in itself is uninfluenced and the contrast medium is situated in the tubular cells and lumina. If the intrapelvic pressure is rising at the beginning of the urography a part of the contrast medium has had time to be excreted into the ducts through the cells and glomeruli; if the intrapelvic pressure

already counterbalances the filtration pressure when the urography is started, a part of the contrast medium has probably reached the proximal parts of the ducts from the blood through osmotic filtration by the glomeruli. However, since the nephrographic effect increased despite the reduction of filtration pressure a great part of the nephrographic effect must have been caused by the accumulation of the contrast medium in the tubular cells.

In the second series the PAH reduced the tubular transport of the iodine compound, while the conditions for the glomerular excretion were unchanged. The poor nephrographic effect thus appears to depend upon altered tubular cell function.

The difference in density of the contrast-filled left pelvis with and without PAH injection is also an indication of altered contrast medium excretion. A study of the density of the right pelvis and ureters demonstrates a similar difference between the two series, the urine having a high concentration of the iodine compound in the series without the depressant.

In the first series a nephrographic effect in both kidneys was visible in all animals after the counter-pressure was released and the glomerular filtration pressure was reduced by blood pressure fall. In the second series, despite the depressant (PAH) two of the seven animals showed a bilateral nephrographic effect. This is explained by the fact that the time interval between the PAH injections and the lowering of the blood pressure in these two rabbits was longer than in the 5 animals having no nephrographic effect, being 40 to 50 min. as compared to 30 min. or less. The excretion of the majority of the PAH is completed in half an hour; so its depressive effect is no longer active after this period.

Our conclusions, based on the above studies of the nephrographic effect, are that the accumulation of the iodine compound is impeded when the excretion of the contrast medium is depressed by a non-iodine compound also excreted by the tubules. In addition, the series demonstrate that the concentration of the contrast medium in the pelves and ureters is lower in the urographies performed with depressants than at those without.

Confirmation of the above findings was provided by a series of clearance determinations in dogs in which urography was performed simultaneously with constant intravenous PAH infusion. In these animals both kidneys showed contrast medium excretion of low concentration into the pelves as well as absence of nephrographic effect after the deliberate lowering of the blood pressure.

It seems to us that the observations may explain variations in the nephrographic effect produced by ureteral occlusion in human beings. In some cases it is intense and in others of a low degree. It may depend

upon the duration of the stasis; if the stasis has been of a long duration, substances from the metabolism of the body excreted via the tubular route (creatinine, hippuric acid) may have occupied the tubular cells. When urography is performed these substances may then depress the excretion of the iodine compound with a resultant low nephrographic effect.

With regard to the present day widespread use of the penicillin therapy we would emphasize that penicillin, due to its excretion via the tubular route, depresses the contrast medium excretion in urography. This must be considered in the differential diagnosis when a low density of the contrast urine is encountered.

## SUMMARY

An investigation of urography in rabbits demonstrated that the nephrographic effect is reduced when the tubular excretion of the iodine-containing contrast medium is depressed by a non-iodine compound also excreted via the tubular route. Furthermore the concentration of the contrast medium in the pelves and ureters is similarly reduced. The mechanism of the depressed tubular excretion in its application to practical work is briefly discussed.

## ZUSAMMENFASSUNG

Eine urographische Untersuchung an Kaninchen zeigte, dass der nephrographische Effekt reduziert wird, wenn die tubuläre Exkretion des jodhaltigen Kontrastmittels durch eine nicht jodhaltige, ebenfalls über die Tubuli ausgeschiedene Verbindung, herabgesetzt wird. Weiterhin wird die Konzentration des Kontrastmittels in den Nierenbecken und Ureteren in ähnlicher Weise reduziert. Der Mechanismus der herabgesetzten tubulären Ausscheidung in Bezug auf die praktische Arbeit wird kurz diskutiert.

## RÉSUMÉ

Une étude expérimentale de l'urographie sur des lapins a montré que l'effet néphrographique est diminué quand l'excrétion tubulaire du moyen de contraste iodé est réduite par un composé non-iodé, excrété lui aussi par la voie tubulaire. De plus, la concentration du moyen de contraste est pareillement réduite dans les bassinets et les uretères. Les auteurs étudient brièvement le mécanisme de cette réduction de l'excrétion tubulaire dans son application au travail pratique.

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# BRONCHIECTASIS OF THE APICAL SEGMENT OF THE LOWER LOBE

by

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Bronchiectasis of unspecific origin is usually reported as localized to the lower lobes of the lungs especially the left. It should be pointed out, however, that the apical segments often remain unaffected; in addition it is the middle lobe on the right side and the corresponding region on the left (the lingula) which are generally involved. A very common combination of sites is a lower lobe with the right middle lobe or the lingula segment of the left upper lobe. The upper lobe bronchi, except those of the lingula, are rarely affected unless there is a tuberculous origin of the disease.

BROCK has shown that bronchiectasis of the middle lobe often has a tuberculous background, *e. g.* the condition usually follows atelectasis caused by enlarged tuberculous glands; this also occurs in other regions of the lungs, without shrinking of the parenchyma necessarily being the direct cause of the bronchiectasis. In 30 to 40 % of all cases, bronchiectasis is found to be bilateral. As regards its frequency in the sexes DUKEN and VON DEN STEINEN have drawn the conclusion, partly from the literature and partly from their own cases, that bronchiectasis in the young is more frequent in the female, contrary to that which obtains in adults. It is, however, difficult to find in the literature a detailed description of the distribution of bronchiectasis based upon reliable modern bronchographic techniques.

A material of 100 cases examined at our hospital during the period from 1950 to 1955 has been carefully analysed (Table). Bilateral bronchography was performed in all cases. The investigation confirms in the main the established opinion, although several interesting points arise.

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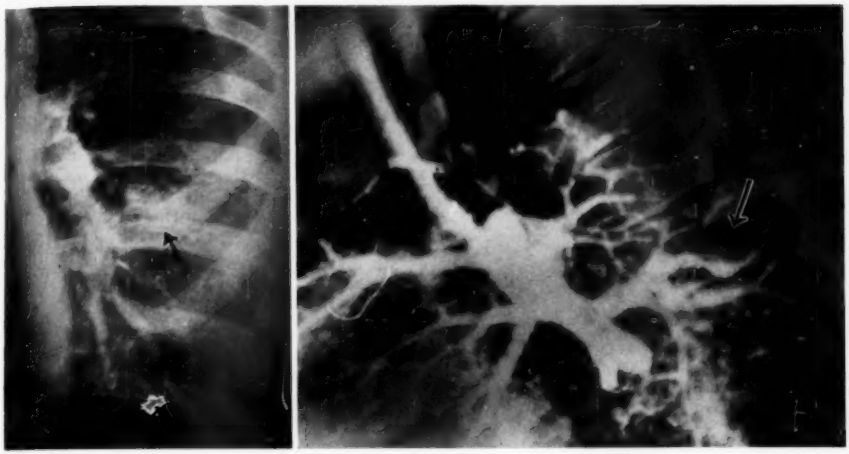
**Table**

*Distribution of bronchiectasis in 100 cases examined by bronchography on both sides*

| | Bilateral 46 | | Unilateral 54 | |
|---|---|---|---|---|
| | Right | Left | Right 21 | Left 33 |
| Lower lobe, basal segments | 22 | | 10 | |
| Lower lobe, basal + apical segments | 17 | | 4 | |
| Lower lobe, apical segment | 2 | | 3 | |
| Lower lobe + middle lobe | 27 | | 8 | |
| Lower lobe + middle lobe + upper lobe | 6 | | 3 | |
| Lower lobe + upper lobe | | | 1 | |
| Middle lobe | 9 | | 3 | |
| Middle lobe + upper lobe | 3 | | 1 | |
| Upper lobe | | | 3 | |
| Lower lobe, basal segments | | 39 | | 24 |
| Lower lobe, basal + apical segments | | 14 | | 18 |
| Lower lobe, apical segment | | 1 | | 1 |
| Lower lobe + lingula | | 28 | | 11 |
| Lower lobe + lingula + apicoposterior segment of upper lobe | | 9 | | 3 |
| Lower lobe + apicoposterior segment of upper lobe | | 1 | | 1 |
| Lingula | | 4 | | 3 |
| Lingula + apicoposterior segment of upper lobe | | 1 | | |
| Apicoposterior segment of upper lobe | | 1 | | |

Thus, the percentage of bilateral ectases has been as high as 46 %, probably due to the improved technique of bronchography. Isolated ectases of the middle lobe — without combination with ectases in other regions — is not very common, having a frequency of 12 %, and when the lingula is alone involved, of only 7 %. There is verification of the accepted notion that well-drained bronchi are less likely to be the site of bronchiectasis. Ectases in an upper lobe alone should always be suspected as having a specific genesis. The material consisted mostly of adults and only 6 were less than 15 years old; the youngest patient was 9 and the oldest 73 years of age. Any difference in the distribution between the sexes could not be shown; 50 cases were males and 50 females. As already mentioned bronchiectasis of the lower lobe is usually confined to the basal segment with the apical segment less often affected. In this material ectases of the lower lobe occurred in a total of 85 %, with the apical segment unaffected in 32 %. Ectases isolated to an apical segment is an unusual finding; in combination with ectases in other regions of the



a b

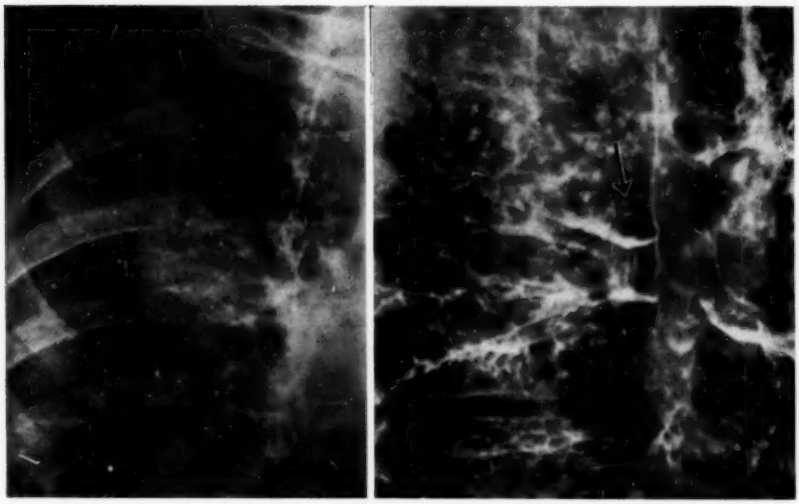
Fig. 1. Case 1. a) Ill-defined opacities on left side. b) Saccular bronchiectases at same site in apical segment of lower lobe.

other lung (which dominated the picture) it was found in 2 cases. Unilateral ectases localized exclusively to an apical segment of the lower lobe occurred in this material in 4 cases, or 4 %.

As these are of special interest, they are included in the case reports together with two more (Cases 2 and 6) which, as they were not examined on both sides, are not included in the material in the table.

*Case 1.* Female, born 1908. Since 1945 repeated haemoptyses. No expectoration. Bronchoscopy 1950 and 1951 showed no changes. At roentgen examination of the lungs 1951 a rounded ill-defined opacity was found in the apical segment of the left lower lobe (Fig. 1 a). One month later only a few fibrous streaks were noted in the region of the former opacity. At bronchography a few days later, saccular ectases were seen in the same region (Fig. 1 b). The remaining bronchi of the left lung were normal. Bronchography of the right lung showed no changes. Resection of the apical segment of the left lower lobe was performed. Histologic examination showed cylindric ectases with very little inflammatory reaction.

*Case 2.* Male, born 1896. Since 1927 repeated haemoptyses. Roentgen examination 1952 demonstrated a rounded ill-defined opacity of the parenchyma of the apical segment of the right lower lobe (Fig. 2 a) and at bronchoscopy bleeding from this area was observed. One month later only a slight streaky opacity remained in the region. At bronchography of the right side bronchiectatic dilatation of a superior branch of the apical segment bronchus of the lower lobe was found (Fig. 2 b). Tb culture negative. As the patient later developed a cough with expectoration, a further bronchography was carried out two years later with the same result. Nine months later the opacity in the apical segment of the lower lobe had almost entirely disappeared.



a b

Fig. 2. Case 2. a) Ill-defined opacity of parenchyma on right side. b) Dilatation of branch of bronchus of apical segment of lower lobe.

*Case 3.* Male, born 1916. 1942 haemoptysis. Roentgen examination of the lungs 1956 (in Hungary) is said to have shown 'veiling of the right lower lobe'. Fresh haemoptysis in May 1954 but roentgen examination revealed nothing abnormal. At bronchography of the right lung several saccular ectases were observed in the apical segment of the lower lobe (Fig. 3). The other branches of the right bronchial tree were normal; bronchography of the left lung normal.

*Case 4.* Male, born 1923. 1940: occasionally brought up blood. Roentgen examination 1949 revealed an oval opacity in the apical region of the left lower lobe. Further haemoptyses 1950 and 1952. At bronchoscopy bleeding from the left main bronchus was observed. At bronchography of the left lung, ectases in lateral branches of the lower lobe were found but no change in the other bronchi. Further haemoptysis in February 1954. The opacity in the left lung had disappeared at roentgen examination two months later. At bronchography of the left lung it was found, however, that the ectases in the apical segment of the lower lobe had increased. Bronchography of the right lung showed no changes. Resection of the apical segment of the left lower lobe was performed. Histologic examination: Fairly definite signs of chronic inflammation. No evidence of tuberculosis.

*Case 5.* Male, born 1908. Spring 1955: repeated minor haemoptyses. At roentgen examination no changes other than old pleurisy on left side. Bronchography of the right side showed nothing abnormal but on the left side there were saccular and cylindric ectases in the superior bronchus of the apical segment of the lower lobe (Fig. 4). Resection of the apical segment was performed. Histologic examination: Bronchiectasis with nonspecific inflammation peribronchially and in the mucous membranes.

*Case 6.* Female, born 1901. Transitory infection of the upper respiratory tract at beginning of September 1954. At the end of the same month repeated haemoptyses

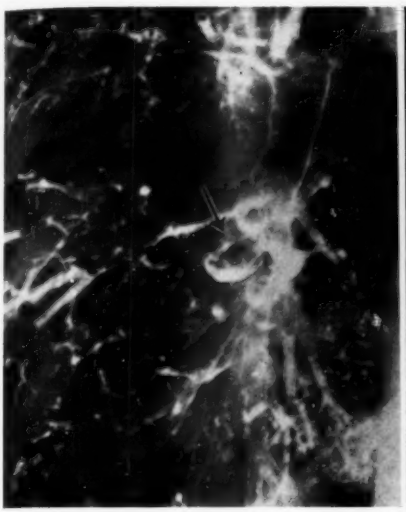


Fig. 3.

Fig. 4.

Fig. 3. Case 3. Saccular bronchiectases of apical segment of right lower lobe.
Fig. 4. Case 5. Saccular and cylindric bronchiectases of apical segment of left lower lobe.

with a homogeneous opacity in the left upper lobe at roentgen examination. Five days later considerable atelectasis of the left upper lobe and a small pneumothorax were found (Fig. 5 a). Two days later the atelectasis had nearly completely disappeared. At bronchography 1955, saccular ectases were present in the lower bronchi of the apical segment of the left lower lobe. The anterior bronchus of the upper lobe was obliterated (Fig. 5 b). At further bronchography five months later this obliteration had disappeared and even the anterior bronchus of the upper lobe filled. With the exception of the formerly observed ectases in the apical segment of the lower lobe no changes could be found. Tb culture negative. Bronchography of the right side was not performed.

## Discussion

All these cases were manifested by haemoptyses. Only in one (Case 2) was there expectoration; the others had no other signs. Cases 1, 2 and 4 had rounded ill-defined opacities of the parenchyma in the apical part of the lower lobe apparently caused by collections of blood in the ectases. Possibly identical changes had taken place in Case 3 but unfortunately the films were not available for examination. In Case 5 changes in the parenchyma could not be proved. Case 6 differs from the rest as to the site of the collection of blood. Coagula had been coughed up from the bleeding area to the upper lobe, blocking the anterior branches of the upper lobe and resulting in massive atelectasis.

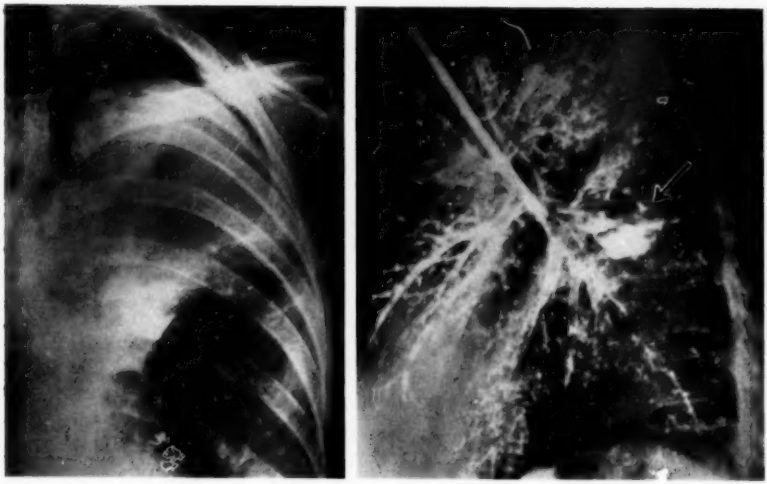


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Fig. 5. Case 6. a) Atelectasis of left upper lobe and pneumothorax. b) Ectases of lower bronchi of apical segment of left lower lobe. Anterior bronchus of upper lobe obliterated.

The apical part of the lower lobe is a common site for tuberculosis, and carcinoma of the lung often arises in this segment. The cases now published show that opacities in this area associated with a history of haemoptysis may indicate localized bronchiectasis. If these opacities disappear and return at a later date, the diagnosis is confirmed. As may occur with minor ectases of the bronchi in other parts of the lung, it may happen that no abnormal signs are found on routine roentgen examination. As in Case 6, bleeding from ectases may even cause large areas of atelectasis, and then the probability of a tumour increases. Bronchography offers a definite diagnosis in all these cases but it ought to be mentioned that the examination, when bronchiectasis is suspected, should always be performed in such a way that all bronchial branches are filled as far as the alveoli, otherwise small peripherally located ectases may remain unobserved. Concerning the genesis of these ectases it may be concluded that no evidence of their association with tuberculosis has been recorded. The histories never contain characteristic features and it is probable that the ectases, as bronchiectasis in general, may be traced back to early childhood.

## SUMMARY

In a material of 100 cases of bronchiectasis in which bronchography of both lungs had been performed, earlier opinions on the localization of ectases were largely confirmed. Isolated ectases in an apical segment of the lower lobe were found in 4 % of cases. These cases had so called dry bronchiectases, disclosed by haemoptyses. The bleeding can result in rounded ill-defined opacities in the apical region of the lower lobe similar to the changes caused by tuberculosis and peripheral carcinoma. The correct diagnosis is confirmed by the disappearance of the opacities and their return at a later date. Bronchography should always be performed.

## ZUSAMMENFASSUNG

In einem Material von 100 Bronchiektasiefällen, in denen eine Bronchographie beider Lungen ausgeführt wurde, ist die frühere Auffassung über die Lokalisation der Ektasien weitgehend bestätigt worden. Isolierte Ektasien in einem apikalen Abschnitt des Unterlappens wurde in 4 % aller Fälle gefunden. Diese Fälle hatten sogen. trockene Bronchiektasien, welche durch Haemoptoen verschlossen waren. Die Blutung kann runde schlecht begrenzte Verdichtungen in der apikalen Gegend des Unterlappens erzeugen, welche den durch Tuberkulose und peripheres Karzinom hervorgerufenen Veränderungen ähneln. Die korrekte Diagnose wird durch das Verschwinden der Verdichtungen und deren späteres Wiederauftreten bestätigt. Eine Bronchographie sollte stets vorgenommen werden.

## RÉSUMÉ

L'étude d'une série de 100 cas de bronchectasies où on avait pratiqué une bronchographie bilatérale a amplement confirmé les idées antérieures sur la localisation des ectasies. L'auteur a trouvé des ectasies isolées dans un segment apical du lobe inférieur dans 4 % des cas. Ces cas présentaient des bronchectasies dites sèches, révélées par des hémoptysies. L'hémoptysie peut donner lieu à des opacités arrondies mal limitées dans la région apicale du lobe inférieur, comparables aux aspects dûs à la tuberculose et au cancer périphérique. Le diagnostic exact est confirmé par la disparition des opacités et par leur réapparition ultérieure. La bronchographie devrait être toujours pratiquée.

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# A SIMPLE INDICATOR FOR THE UTERINE CERVIX IN OBSTETRIC ROENTGEN EXAMINATIONS

by

*Stig Samuelsson* and *Alf Sjövall*

The diagnosis of first and second degrees of placenta praevia in soft tissue films as well as in arteriograms depends on a rather crude estimate of the distance between the margin of the placenta and some arbitrary point on the film, where the internal os of the cervix is supposed to lie. Even in cases of complete placenta praevia (third degree), in which only a flap or lobe of the placenta covers the os, the interpretation of the film may be difficult. In order to obtain more exact information regarding the site of the os, we have tried ordinary hairdressing clips as indicators in a number of cases with bleeding in the last trimester and in which placenta praevia was suspected. A similar method was first described by STALLWORTHY (1951), his original intention being to give visual proof of the distance which could exist between the tip of the examining finger, if it were inserted just inside the os, and the lower pole of a placenta which was situated high in the lower segment.

With clips on the anterior and posterior lips of the cervix and a volsellum in the posterior vaginal vault STALLWORTHY together with REID measured the distance between the os and the brim of the pelvis. The brim approximately corresponds to the junction of the upper and lower uterine segments. The distance was found to be about 7 centimetres. Hence the margin of a placenta praevia of the first degree lies approximately at this distance from the os.

In our cases ordinary hair-dressing clips or hair-pins (Fig. 1) were applied to the cervix before roentgen examination. The clip is after-

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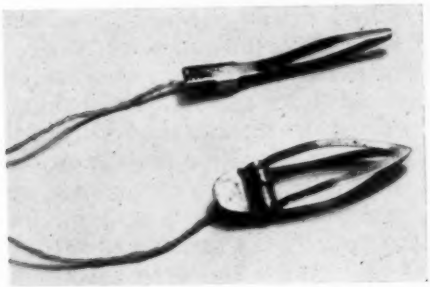

Fig. 1. A clip and a pin with attached threads.

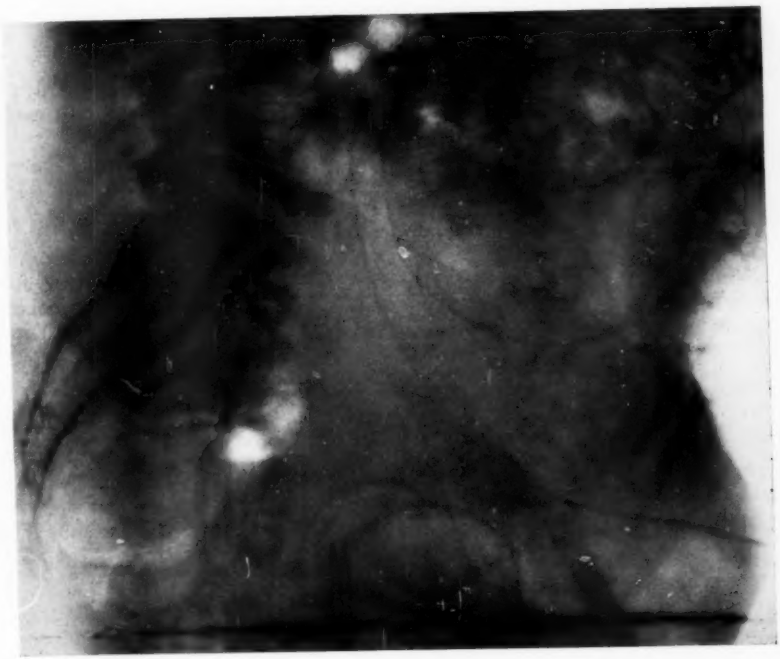

Fig. 2. Angiography. Complete placenta praevia. The distance between the clip and the placental margin in this case was only 3 cm.

wards easily removed by a thread attached to it. Neither the application nor the removal of the clips or pins damages the cervix. In 12 cases of bleeding in the last trimester of pregnancy 8 were judged by soft tissue roentgenograms only and 4 by angiography. In angiographic films the distance between the tip of the clip and the border of the placental sinuses is distinctly seen (Fig. 2); in soft tissue films it can also be measured. In 9 cases where this distance was $\geqq 10$ centimetres the placenta was not praevia. In 3 cases of complete placenta praevia, however, the distance was only 3 to 5 centimetres. Thus a distance of $\geqq 10$ centimetres between the os and the placental border seems to exclude placenta praevia, if conclusions can be drawn from this small series of cases. The tip of the clip does not, of course, correspond exactly with the site of the internal os. The use of clips on the cervix, however, provides a far better localization of the internal os in the roentgen film than a mere estimation of something that is not seen at all.

## SUMMARY

Ordinary hair-dressing clips were used to indicate the site of the uterine cervix in cases in which placenta praevia was suspected. A distance between the clip and the border of the placenta equal to or greater than 10 centimetres seems to exclude placenta praevia.

## ZUSAMMENFASSUNG

Gewöhnliche Haarspangen sind benutzt worden, um die Lage der Cervix uteri in Fällen mit Verdacht auf Placenta praevia zu markieren. Ein Abstand zwischen der Spange und der Kante der Placenta von 10 Zentimeter oder mehr scheint das Vorliegen einer Placenta praevia auszuschliessen.

## RÉSUMÉ

Les auteurs ont utilisé des agrafes à cheveux pour indiquer la situation du col de l'utérus dans des cas où on soupçonnait un placenta praevia. Une distance entre l'agrafe et le bord du placenta égale ou supérieure à 10 cm semble exclure le placenta praevia.

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# TANTALUM POWDER AS AN INDICATOR OF THE BRAIN TUMOUR REGION FOR POSTOPERATIVE RADIOTHERAPY AND THE DIAGNOSIS OF RECURRENCE

by

*M. Lindgren, F. O. Löfgren* and *N. Lundberg*

In 1950 WHITCOMB & SCOVILLE described the use of tantalum powder for the postoperative roentgenographic demonstration of the surfaces of the incision made at open lobotomy. This method was used in over 125 cases without visible harmful effects or any increase in the incidence of epilepsy.

This paper is a report of our experience with tantalum powder used as an indicator in combined surgical and radiologic therapy of glioma in the cerebral hemispheres. The purposes of the method were:

1) to locate the resection surfaces postoperatively and any remaining tumour tissue and thereby to enhance the selectivity of radiotherapy, and

2) to enhance early detection of any recurrence by control roentgenography.

The size of the particles of the tantalum powder (Manufactured by Fansteel Metallurgical Corporation, North Chicago, Illinois) used was 8—12 $\mu$ (325 mesh). The powder was stirred to a thick paste with normal saline. The surface to be demonstrated was then covered with a thin layer of the paste. In some cases the powder was spread over very thin

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slices of oxycel, which were placed on the surface to be indicated. In a few cases in which the tumour was verified by diagnostic puncture a small amount of tantalum paste was deposited via the cannula at the site from which aspirated material had contained tumour tissue.

In 25 cases the method was used as a guide for postoperative radiotheraphy. Radiotheraphy was planned on the basis of the preoperative roentgen examination in combination with the postoperative roentgenogram in which the tantalum powder indicated the site of any remaining tumour tissue. This facilitated focussing of the roentgen beam onto the tumour region with the minimal irradiation of surrounding healthy tissue.

Thirteen patients were repeatedly examined roentgenographically for recurrences. Migration of the tantalum powder suggesting recurrence was demonstrated in 10. These patients had been followed up for 3 months to 1 year and 6 months. In 3 of the cases in which the tumour was situated in the frontal region the recurrence was verified encephalographically and at reoperation. In the other 7 cases (2 frontal, 1 parietal, 4 temporal) the recurrence was confirmed by the further course of the disease. In 4 cases displacement of the tantalum powder was shown before the appearance of any clinical signs of recurrence. As examples of tumour recurrence Cases 1 and 2 are described in detail in the case reports below.

In 3 cases no migration of the tantalum powder suggestive of a recurrence was demonstrable. These patients were followed up for 3 years to 4 years and 9 months. None of the patients showed clinical signs of a recurrence. Case 3 is an example of cases without tumour recurrence.

## Case reports

*Case 1.* A man, aged 32. Operation revealed a large tumour laterally in the frontoparietal region. The frontal lobe was resected through the middle of the tumour. The resection surface was covered with a layer of tantalum powder. Microscopic examination showed astrocytoma. The patient did not receive radiotherapy after operation.

The first roentgen examination 1 month after operation showed an irregular strip of tantalum powder half a finger-breadth wide in the lateral upper part of the resection surface (Fig. 1). Control roentgenography revealed no further changes until five and a half months after operation, when the strip of tantalum powder was slightly curved, displaced medially in the frontal view, and spread out especially anteriorly and inferiorly in the lateral view (Fig. 2). This migration of the tantalum powder indicated a recurrence although no clinical signs of this were observed. One year after the operation the patient complained of transient weakness of the left arm. After a further 6 months he had obvious symptoms of a recurrence in the form of weakness and paresthesia of the left arm and hand, frontal headache with nausea and increasing apathy. Roentgen examination in the meantime had shown that the tantalum powder was now markedly increased in width in both the frontal and lateral views (Fig. 3).

Encephalography demonstrated that the right anterior horn of the lateral ventricle was pushed entirely under the falx. Its lateral wall coincided with the medial border of

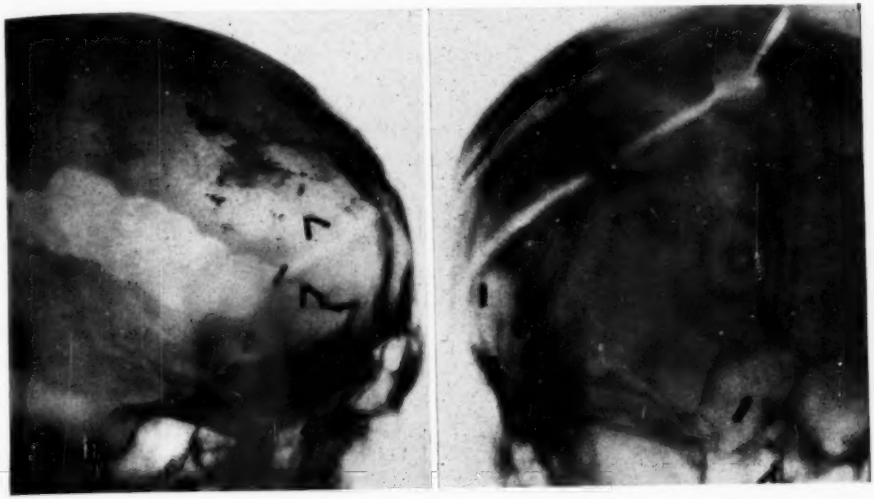

Fig. 1. Case 1. One month after frontal resection. Small amount of tantalum powder in the lateral upper part of resection surface.

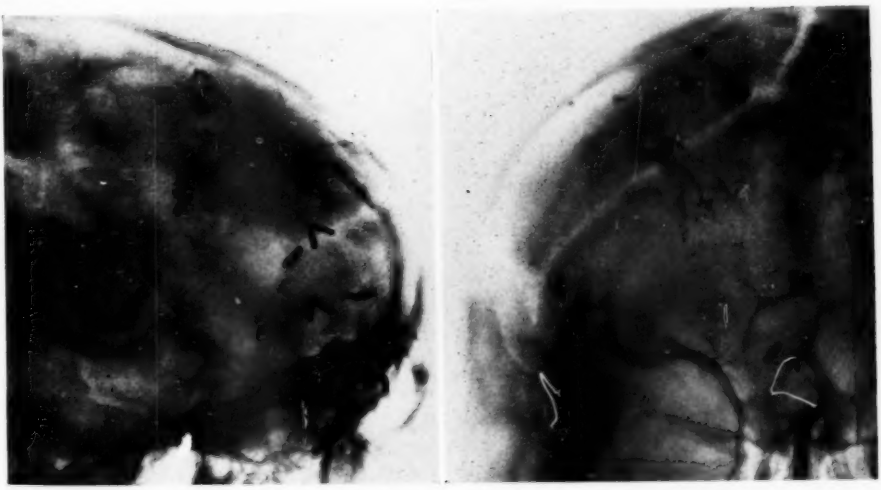

Fig. 2. Case 1. Five and a half months after operation. Lateral view: Separation of tantalum powder especially anteriorly and inferiorly. Frontal view: Tantalum powder slightly curved and displaced medially. Recurrence. No clinical signs.

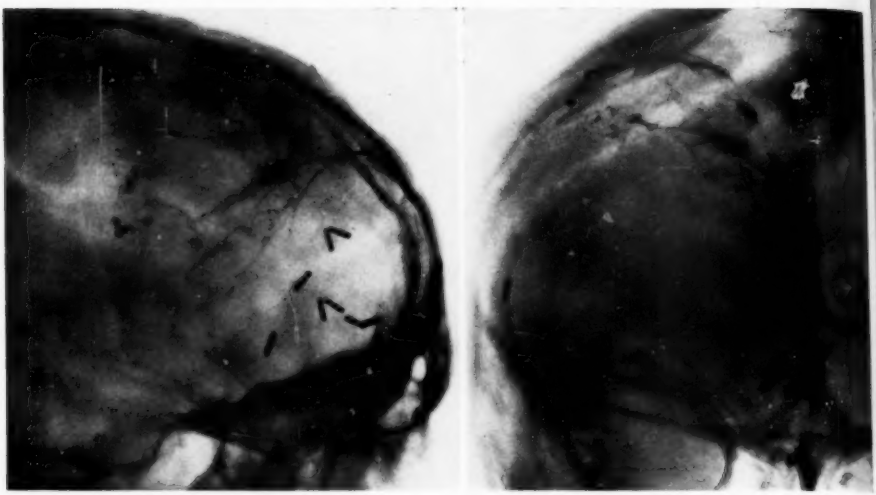

Fig. 3. Case 1. One year and six months after operation. Spread of tantalum powder markedly increased in both frontal and lateral views. Obvious clinical signs of recurrence.

the tantalum powder. At reoperation the site of the previous tumour was found to be occupied by a tumour the size of an orange. Half of the tumour was embedded in the temporal lobe.

In this case displacement of the tantalum powder in the roentgenogram showed a recurrence 6 months before the appearance of clinical signs. The growth of the tumour would then be followed during the subsequent year until reoperation.

*Case 2.* A woman, aged 35. Operation revealed a tumour in the right temporal lobe growing centrally towards the insular region. The temporal lobe was resected and all visible tumour tissue was removed; tantalum powder was placed in those parts where residual tumour tissue might possibly still be present. Microscopic examination showed astrocytoma. After operation the patient received roentgen therapy.

On roentgen examination 2 weeks after the operation the tantalum powder was seen in the anterior part of the temporal lobe in a region 3 cm in diameter. Its medial border lay 2 cm from the midline and its upper border 4.5 cm above the floor of the middle cranial fossa (Fig. 4).

Two and a half years after operation hemiparesis rapidly occurred and the patient became mentally blunt. Roentgenograms taken a few months later showed that the tantalum powder had been displaced 2 cm downwards: its upper outline, which was formerly convex upwards was now concave (Fig. 5). This downward displacement and the change in the shape of the tantalum deposit indicated a recurrence. The patient received no further treatment and gradually deteriorated with signs of increasing intracranial pressure.

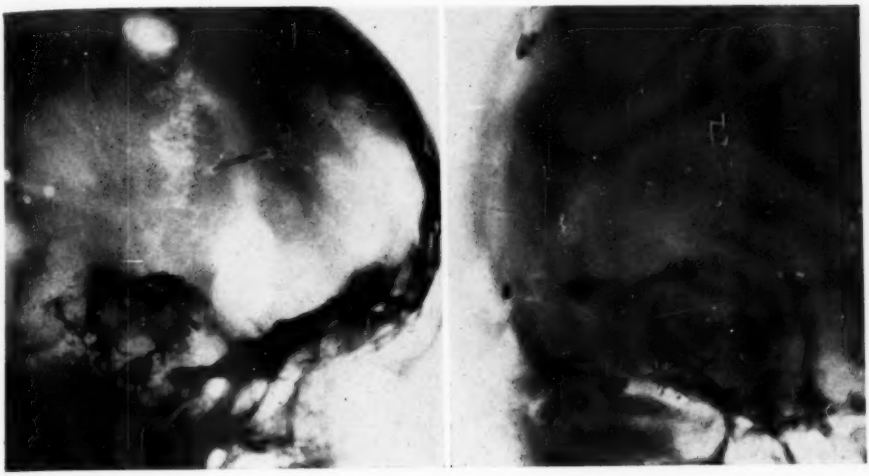

Fig. 4. Case 2. Two weeks after resection of temporal lobe. Tantalum powder on resection surface; upper outline 3 cm above floor of middle cerebral fossa, convex upwards.

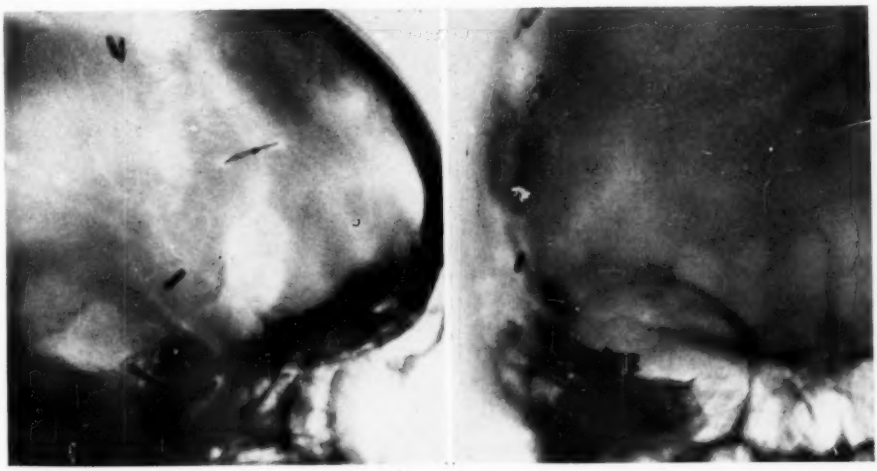

Fig. 5. Case 2. Two and a half years after operation. Tantalum powder pushed markedly forwards and downwards; upper outline now concave. Recurrence.

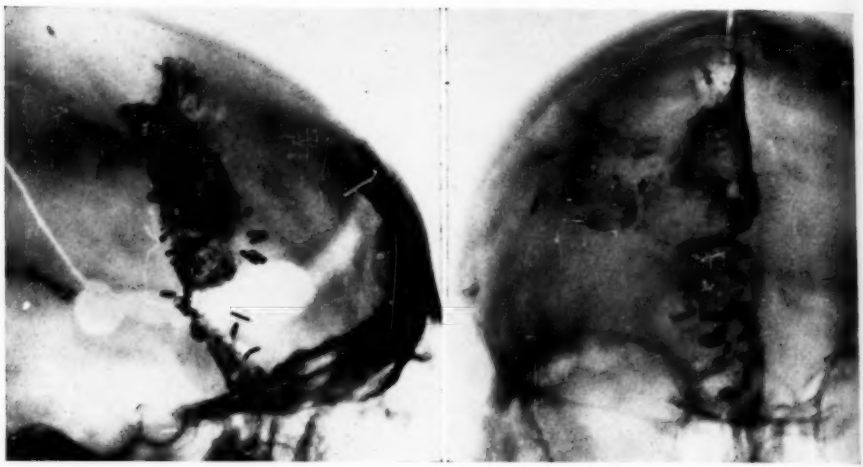

Fig. 6. Case 3. Two weeks after frontal resection. Tantalum powder covering medial half of resection surface.

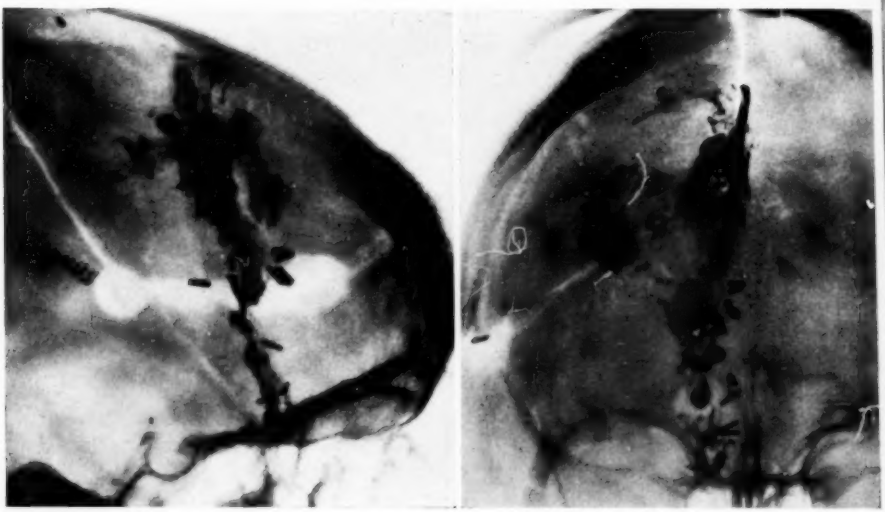

Fig. 7. Case 3. Three months after operation. Tantalum powder displaced 1 cm forwards. Displacement of tantalum powder 1 cm to right below falx. Postoperative shrinkage of brain.

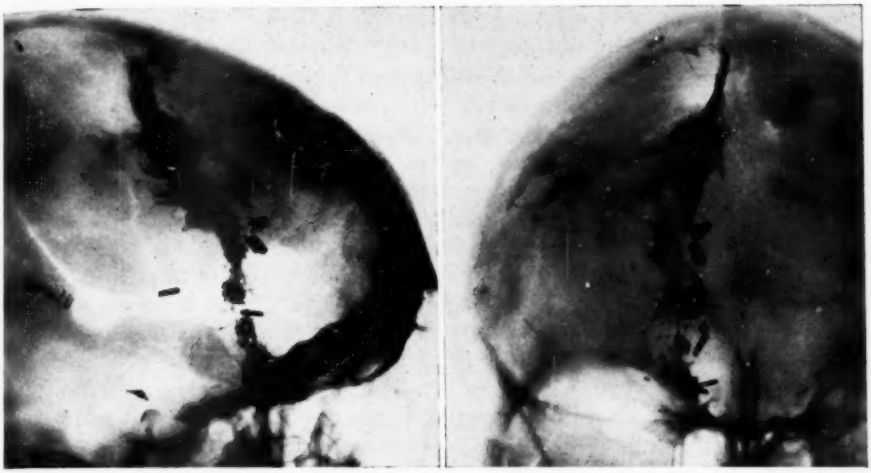

Fig. 8. Case 3. Four years and nine months after operation. No further change in position of tantalum powder. No recurrence.

*Case 3.* A woman, aged 43. Operation revealed a tumour located in the medial part of the right frontal lobe. The frontal lobe was resected with the upper part of the resection surface in the coronary plane and the lower part just before the ala parva. All visible tumour tissue was removed. The resection surface was covered with tantalum powder. Microscopic examination showed astrocytoma with signs suggestive of malignancy. The patient did not receive radiotherapy after operation.

Roentgen examination 2 weeks after operation showed tantalum powder covering the medial half of the resection surface and extending to the midline (Fig. 6). Roentgen examination 3 months after operation: Below the falx the tantalum powder was now displaced to the right with the medial outline 1 cm to the right of the midline. The entire layer of tantalum powder was displaced 1 cm forwards (Fig. 7). Since then control roentgenography — the last examination 4 years and 9 months after the operation — has shown no further change in the position of the powder (Fig. 8). On encephalography two years and a half after operation the pellucid septum was displaced almost 1 cm to the right, coinciding with the medial outline of the tantalum powder. During the follow-up period the patient has shown no symptoms or signs of a recurrence.

The displacement of the tantalum powder observed 3 months after the operation was interpreted as the result of the healing process during the postoperative stage. This assumption was supported by encephalography two and a half years later which showed only signs of shrinkage of the frontal lobe. This suggests that roentgenograms should be taken immediately after operation and then again about 2 months later, by when the postoperative change of the cerebrum has presumably ceased.

Later roentgenograms should then be compared with the films taken at the second examination in order to judge whether any migration of the tantalum powder indicates a recurrence.

## Discussion

It is known that even prolonged contact between the metal tantalum and body tissues will produce hardly any reaction. Histologic studies by BAILEY, INGRAHAM, WEADON & SUSEN showed that this also holds for tantalum powder in contact with brain tissue. We were able to confirm this in one case in which histologic examination post mortem showed only slight inflammatory reaction with scanty cellular infiltration round the tantalum particles. Like WHITCOMB & SCOVILLE, we observed no clinical signs suggesting any undesirable side-effects even when fairly large amounts of tantalum powder were deposited directly onto the resection surface of the brain.

The amount of tantalum powder used varied from one case to another. For detection of any recurrence it proved advantageous to spread the powder generously over the entire resection surface.

Migration of the powder due to recurrence varied. The entire tantalum powder was sometimes displaced without any appreciable change in shape, in other cases the tantalum powder was spread by infiltration of tumour tissue, and occasionally it was curved around the expansive process.

These changes can, of course, be judged more readily if the entire resection surface is covered with tantalum than if the powder is deposited in the form of small masses in a few sites. Another way of indicating the resection surface is to use silver clips. But like small deposits of tantalum powder, clips are less informative than powder spread all over the surface.

Recognition of any migration of the powder requires that the roentgenograms be taken with the same projection at every examination. The examination should preferably include not only frontal and lateral but also axial films, which provide good views of the frequently oblique resection surfaces.

As mentioned in connection with Case 3 the roentgenograms should be taken as soon as possible after the operation and then again 2 months later, by when migration of the tantalum powder due to sedimentation and postoperative changes of the cerebrum have probably ceased. Any subsequent displacement of the tantalum powder suggests a recurrence.

As is apparent from the case reports, the method provides a possibility of demonstrating recurrences before the appearance of clinical signs. The value of the early diagnosis of recurrences of glioma is questionable.

As a rule neither surgical nor roentgenologic treatment are indicated by recurrence and an early diagnosis is only of prognostic value. Occasionally, however, the early diagnosis of a recurrence can be of therapeutic value, such as in slowly growing frontal or occipital gliomas treated by resection of the lobe. In these cases we do not consider radiotherapy indicated until a recurrence has been demonstrated and even then sometimes not until after reoperation; the early diagnosis of a recurrence is consequently of value.

## SUMMARY

Tantalum powder has been applied at operation to the resection surface of gliomas of the cerebral hemispheres or, in cases in which only a diagnostic puncture has been made, the tantalum powder has been injected at the site of the puncture. In planning radiotherapy, the indication of the tumour site in the post-operative roentgenograms by the tantalum has proved of value in the focussing of the roentgen beam. Signs of displacement of the tantalum powder at control roentgenography provide a means of making an early diagnosis of a recurrence.

## ZUSAMMENFASSUNG

Tantalpulver ist während der Operation auf die Resektionsoberfläche von Gliomen der Grosshirnhemisphären appliziert worden. In Fällen, bei denen nur eine diagnostische Punktion gemacht wurde, ist Tantalpulver in die Punktionsstelle injiziert worden. Bei der Planlegung der Strahlenbehandlung hat sich die Tantalmarkierung des Tumorgebietes auf postoperativen Röntgenbildern für die Fokussierung der Röntgenstrahlen als wertvoll erwiesen. Lageveränderungen des Tantalpulvers auf Kontrollaufnahmen erlauben die Stellung der Frühdiagnose eines Rezidives.

## RÉSUMÉ

Les auteurs ont déposé de la poudre de tantale, au cours de l'opération, sur la surface d'exérèse de gliomes des hémisphères cérébraux, ou, quand ils n'avaient fait qu'une ponction exploratrice, ont injecté de la poudre de tantale au lieu de ponction. Le repérage du siège de la tumeur par le tantale sur les radiographies post-opératoires s'est montré utile pour le centrage roentgenthérapique. Les signes de déplacement de la poudre de tantale sur les radiographies de contrôle fournissent un moyen de diagnostic précoce des récidives.

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# THE INFLUENCE OF OXYGEN ON THE RADIOSENSITIVITY OF MAMMALIAN TISSUES

by

*E. A. Wright and P. Howard-Flanders*

The decrease in radiosensitivity of human skin caused by ischaemia was first demonstrated by SCHWARTZ in 1909. Using roentgen and gamma-rays he showed that firm pressure applied to the skin during irradiation greatly reduced the subsequent reaction as compared to irradiation under normal conditions. Later JOLLY (1924) found reduced damage in the guinea pig and rat thymus irradiated while the blood supply was occluded by a ligature, and MOTTRAM (1924, 1932, and 1936) observed increased resistance to radiations by occluding the blood supply of the rat's tail by ligature and pressure and when the animals were made anaemic by bleeding; he also observed that tar warts in mice treated with gamma-rays of radium often showed more damage in the deepest parts of the tumours although the more superficial parts received the larger dose. CRABTREE and CRAMER (1933) irradiated thin slices of mouse tumour *in vitro* under aerobic and anaerobic conditions and found that anaerobiosis afforded some degree of protection as judged by subsequent growth in mice.

EVANS et coll. (1942), LACASSAGNE (1942) and DOWDY et coll. (1950) also found a decreased radiosensitivity with various degrees of anoxia. DOWDY's animals were approximately twice as resistant to the effects of radiation when breathing 5 to 7 % oxygen as were those irradiated while breathing air. SCOTT (in a paper by GRAY et coll. 1953) records a

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factor of 1.5 to 2 in increased radiosensitivity of a transplanted mouse tumour when the animals were breathing oxygen at 3 atmospheres compared with the radiosensitivity of the same tumour when the animals were breathing air. Quantitative results for a variety of oxygen tensions have been published for ascites tumour cells (CONGER 1956) and for normal lymphocytes (TROWELL 1953). However, we have found no report of quantitative data on radiosensitivity at a range of oxygen tensions from anoxia to 3 atmospheres' pressure of oxygen, for any intact mammalian tissue. We have attempted to obtain such data for one normal mammalian tissue. For this work we selected the growing bone in the tail of the young mouse.

*Methods.* In all our experiments, we irradiated the tails of seven-day-old mice and measured the amount of growth in the subsequent three weeks. The tail lengths were measured by slipping a graduated glass tube over the tail and pressing it gently against the body of the animal. The growth of the irradiated tail was compared with that of litter-mate controls. Part of the data was obtained from inbred C 57 Black and part from Swiss white mice.

*Control of oxygen tension.* When anoxia was required the tail was inserted into a plastic cuff lined with a thin rubber membrane which could be inflated to a pressure of 300 mm Hg. The pressure blanched the tail and the residual oxygen in the tissues was rapidly metabolised. To check that the tails were made anoxic by the pressure cuff, the radiosensitivity of the tails of two groups of mice was compared. In the first group the tails were compressed; in the second group the whole animal was made anoxic by nitrogen asphyxia for 2 minutes. No difference was found between the two groups in the sensitivity to a fast electron irradiation of about 3,000 rads delivered in one second. Where other concentrations of oxygen were required the animals were made to breathe various partial pressures of oxygen. These were 380 mm Hg of air (considered to be 10 % oxygen), air (20 %), air at 2 atmospheres (40 %), oxygen at one atmosphere (100 %) and oxygen at 3 atmospheres (300 %). None of these procedures carried out alone affected the growth of control animals.

*Irradiations.* The roentgen irradiations were carried out at 190 kV, 6 mA, at a distance of 25 cm. The roentgen rays were filtered by 0.5 mm Cu, 1 mm Al and 12 mm Perspex. The H. V. L. was 1.5 mm Cu and the dose-rate 90 rads per minute. Three different doses were given at each gas pressure. High-intensity irradiations were carried out with 1.6 MeV electrons from a Van de Graaff generator. An approximately uniform dose was given to the tail at a dose-rate of 2,500 rads per second.

*Results.* All the results were calculated on the basis of radiosensitivity,

$$S = \frac{\log_{10} (\text{growth control tails})/(\text{growth irradiated tails})}{\text{dose in kilo rads.}}$$

This was used because the curve relating reduction of growth-rate to dose was approximately exponential. To obtain a mean value of S for the roentgen irradiations when several doses were used, the individual values of S were weighted in inverse proportion to the square of the standard error. A summary of the results are shown in Diagram 1 as a plot of the relative radiosensitivity against partial pressure of oxygen respired. These results have been published in a preliminary communication by HOWARD-FLANDERS & WRIGHT (1955).

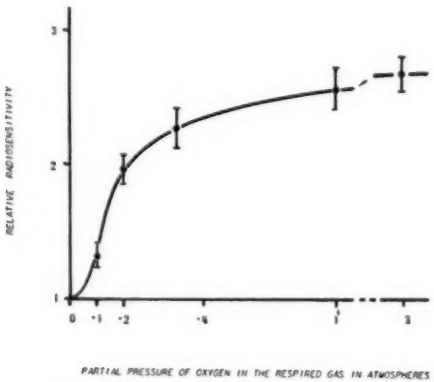


Diagram 1. Curve representing the changes in radiosensitivity obtained in the growing mouse tail with changes in the partial pressure of the respired gas. The lowest radiosensitivity obtained with anoxia produced by pressure was taken as unity.

*Anatomy and histology.* The mice used had 29 tail vertebrae. The first three or four are inside the body and were not measured by our technique. At seven days old the vertebrae vary in length from 0.5 to 1.5 mm, and consist of two ends made up of cartilage (the epiphyseal cartilages) separated by a marrow cavity. The cartilage cells divide and form columns of cells that then enlarge (hypertrophic zone). These cells are then removed by cells adjacent to the marrow and are replaced by trabecular bone. Blood enters the marrow of the bone and travels to each end. Oxygen from the blood diffuses through the hypertrophic zone of cartilage cells to reach the radiosensitive zone of dividing cells, at distance of not less than 80 $\mu$. The normal anatomy of the tail vertebrae is illustrated in Fig. 1.

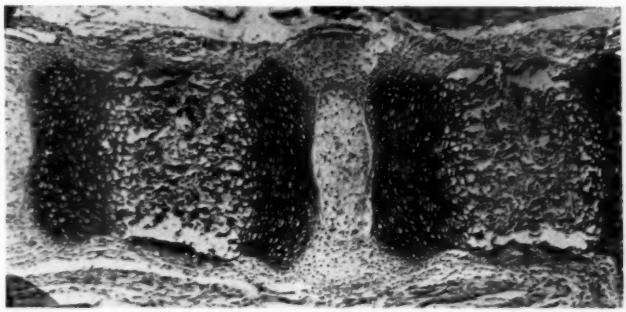

Fig. 1. Two vertebrae of the mid-portion of the tail of a seven day old mouse cut longitudinally. x 50. Stained with haematoxylin and eosin.

## Discussion

The metabolism of oxygen by the intervening hypertrophic cells, combined with the distance which the oxygen must diffuse to reach the dividing cells, results in a reduced supply of oxygen. Thus the oxygen tension normally enjoyed by the cartilage in the dividing cell zone must be well below the oxygen tension of most normal tissues. The slow rise of the curve in Diagram 1, when compared with data obtained for single cell suspensions, is attributed to this cause.

The results are also affected by temperature, which modifies oxygen consumption and at the same time affects the calibre of the blood vessels supplying the tissues. The differing reactions of human skin to cold illustrate that these two factors in oxygen supply and demand may vary. Cold skin may be seen blue and anoxic or red and oxygenated, or it may be white when the degree of oxygenation is not apparent. Our experiments were carried out with the young mice at about room temperature (20° C); the animals were pink. The oxygen tension of the sensitive cells at the moment of irradiation is an important factor in determining the radiosensitivity of a normal tissue, but there is no direct way of measuring the oxygen tension in minute layers of specific cells often buried deep in delicate surroundings. However, we have been able to follow changes in the oxygenation of the intact cartilage cells in the mouse tail, by means of high-speed irradiations with fast electrons. The results of this experiment are shown in Diagram 2. We observed that, following blanching of the tail by pressure, the radiosensitivity falls by a factor 1.8 in the course of 4 seconds. It stops at the reduced value for so long as the blood is occluded. If the circulation is permitted to return, the radiosensitivity rises rather more slowly, reaching the former value within 35 seconds.

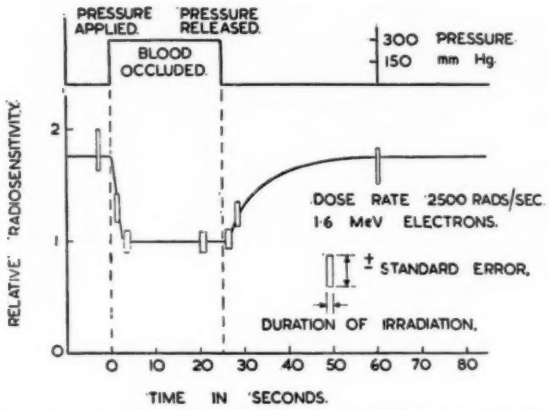


Diagram 2. Curves representing the fall of radiosensitivity following the application of pressure to the tail and subsequent rise in sensitivity with release of pressure.

From published work one can draw two extreme types of oxygen curve for mammalian tissue. On the one hand CONGER has found that mouse ascites cells irradiated *in vitro* and presumably well oxygenated but respiring slowly (his irradiations were done at room temperature and the ascites tumour used has a low $Q_{O_2}$) show a sharp and rapid rise to a high plateau that is reached shortly after the partial pressure of oxygen in air is reached. On the other hand, HOLLCROFT et coll., with a solid mouse tumour, found no difference between when it was irradiated with the blood supply tied or when the animal was breathing air and the tumour was left undisturbed. The continuation of the curve for solid intact tumours is difficult to draw as no strictly comparable data exist, but SCOTT found that there was an increase in sensitivity of 1.5 to 2 when his animals were irradiated when breathing oxygen at 3 atmospheres compared with animals breathing air. Our results for an avascular but normal tissue lie in between these two extremes. It is suggested that these wide differences are due to differences in oxygen transport and consumption, and that if these tissues could be irradiated under conditions of uniform oxygenation the shape of their oxygen-radiosensitivity curves would be more nearly the same. It may well be, however, that although the shapes of the curves might be similar, the steepness of rise or the height of the plateau might differ.

Now that irradiation during the administration of oxygen is being carried out at a number of centres, we wish to draw the attention of radiotherapists to the potential dangers of this type of treatment. It is probable that some normal tissue radiosensitivities may also be increased by the administration of oxygen. Bones have a preferential energy absorption from roentgen irradiation at 250 kV, and bone necrosis fol-

lowing radiotherapy is a well known hazard. Whether there is an increase in radiosensitivity of human bones analogous to our findings in the growing bones of mice remains to be seen, but until this point is established it might be advisable to restrict the total dose delivered when giving oxygen if the field includes bone. The same difficulty does not, of course, occur with supervoltage radiotherapy.

Finally, it is now probable that a great advantage can be obtained by irradiating certain types of tumour during oxygen therapy at high pressure as Dr. CHURCHILL-DAVIDSON has shown. In some tumours, however, greater advantage might be obtained if all the tissues of the body were made anoxic during a short period of irradiation, and if the dose was raised appropriately. We must consider whether both these requirements are obtainable. For example, partial anoxia is frequently obtained during nitrous oxide anaesthesia. Large doses of radiation can be given in extremely short periods of time by the new high-intensity equipment, such as the linear accelerators. Approximately double the normal doses of irradiation would be necessary and would have to be administered in less than two or three minutes.

## SUMMARY

The radiosensitivity of growing bone in the tail of week-old mice has been determined when the tissues were made anoxic by compression and when the animals were breathing partial pressures of oxygen ranging from half an atmosphere of air to three atmospheres of oxygen. When irradiation doses in the form of fast electrons, delivered in approximately one second, were used the tail reached minimum sensitivity following compression in four seconds. The changes in radiosensitivity found under these conditions are discussed in relation to the special anatomy and physiology of the sensitive cartilage cells concerned with the growth of the bones. The relation of these results with other work on mammalian tissue and cells is also discussed and some possible practical applications to radiotherapy are considered.

## ZUSAMMENFASSUNG

Die Radiosensibilität des wachsenden Knochens im Schwanze einer wochenalten Maus ist untersucht worden, nachdem die Gewebe durch Kompression anoxämisch gemacht worden waren und nachdem die Tiere partiellen Sauerstoffdruck atmeten, der zwischen einer halben Atmosphäre Luft und drei Atmosphären Sauerstoff variierte. Bei Verwendung von Strahlendosen in der Form schneller Elektronen, die etwa innerhalb einer Sekunde gegeben wurden, erreichte der Schwanz seine geringste Sensibilität nach einer Kompression von 4 Sekunden. Die unter diesen Bedingungen gefundenen Veränderungen der Radiosensibilität werden in ihrer Beziehung zu der speziellen Anatomie und Physiologie der sensitiven Knorpelzellen, welche mit dem Knochenwachstum zusammenhängen, diskutiert. Die Beziehung dieser Ergebnisse zu anderen Arbeiten über Zellen und Gewebe der Säugetiere wird ebenfalls besprochen und einige praktische Anwendungen in der Radiotherapie werden erwähnt.

## RÉSUMÉ

Les auteurs ont déterminé la radiosensibilité de l'os en croissance dans la queue de souris agées d'une semaine quand les tissus sont rendus anoxiques par compression et quand les animaux respirent des pressions partielles d'oxygène allant d'une demi-atmosphère d'air à trois atmosphères d'oxygène. Irradiée par des électrons rapides pendant environ une seconde, la queue atteint son minimum de sensibilité après compression en quatre secondes. Les auteurs étudient les modifications de radiosensibilité trouvées dans ces conditions en fonction de l'anatomie spéciale et de la physiologie des cellules cartilagineuses sensibles qui interviennent dans la croissance des os. Ils comparent aussi leurs résultats avec d'autres travaux sur les tissus et les cellules des mammifères et envisagent certaines applications pratiques possibles à la radiothérapie.

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# SOME RESULTS OF RADIO-IODINE DIAGNOSTIC TESTS FOR THYROID FUNCTION

by

*D. Brinkley, J. L. Haybittle* and *K. J. Plested*

When iodine 131 was first used for diagnostic purposes in this department, studies were confined to a measurement of maximum percentage uptake and half-life in the thyroid gland. At a later stage the estimation of plasma and protein bound iodine 131 concentration at 48 hours was instituted as a routine test in addition to the uptake measurement. We have now reviewed all the cases which received both types of test during an initial period of 18 months. The objects of our review were to assess the efficacy of each test in the diagnosis of thyroid disorder, and to study in particular the cases where the tests failed so that we might interpret results more accurately in the future. We feel that our analysis has partially achieved these objects, and that some of the points brought out are of sufficient interest and importance to warrant this publication of another series of radioiodine diagnostic tests.

*Physical measurements.* The normal tracer dose was from 70 to 100 $\mu$C of carrier-free iodine 131 administered by mouth. In a few cases where it was required to map the thyroid outline a higher dose was given. In others as a matter of urgency the tracer investigations were carried out using the first drink of a fractionated therapeutic dose, but no results obtained with doses of greater than 500 $\mu$C were included in our series.

Uptake into the gland was measured with a lead cathode GM counter 50 cm from the neck. Two counts were made, one with and one without a lead block 10 × 10 × 5 cm thick shielding the gland. The difference was taken to be due to the activity in the gland, and the technique was

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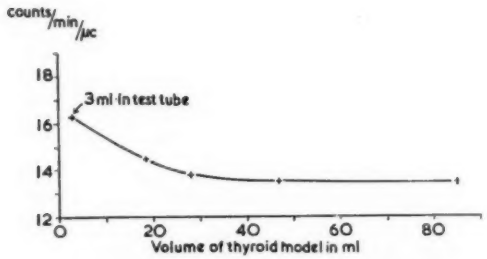


Diagram 1. Calibration curve of counter obtained with hollow model thyroids filled with known amounts of radio-iodine and situated in a water phantom to simulate the neck.

calibrated with known amounts of iodine 131 in four model thyroids having volumes of 18, 28, 47 and 85 cc respectively. Measurements on these models placed in a cylindric water tank simulating the neck region showed that the counts/min/$\mu$C was only a slowly varying function of thyroid volume (Diagram 1). In practice a clinical estimate of gland size was made and the appropriate calibration factor read off from the curve. The uptake was measured 4 to 6 hours after administration of the tracer dose and then daily on four succeeding days. From these points the maximum percentage activity at any time and the effective half-life in the gland were found.

Plasma samples taken at 48 hours were counted in flat bottomed glass tubes resting in contact with the top face of the crystal of a scintillation counter, in a similar manner to that described by Oliver (1953). Separation of the protein-bound portion (which was counted in the same way) was made using the technique described by Goodwin et coll. (1951).

We believe our results are not in error by more than 4 units at the 50 % level of maximum uptake, or by more than 0.03 at the 0.3 % per litre level of protein-bound activity.

*Selection of cases.* All cases referred to this Department for radio-iodine assessment of thyroid function were initially included in our series, except those who were taking anti-thyroid drugs, iodine or thyroid during the test, and those who for the reasons stated earlier received a larger dose than 500 $\mu$C. The cases were the more difficult problems of thyroid disorder found in a general hospital. They included those who had recurrent thyrotoxicosis after surgical removal of the gland, those who had proved unsuitable for treatment with anti-thyroid drugs, cases with auricular fibrillation of uncertain cause, and a group with signs of anxiety and stress and possible mild hyperthyroidism. There were a few cases with unusual types of endocrine imbalance.

All patients had a full history taken and a clinical examination made

by the medical staff of the department before the radio-iodine tests were done. When possible, B.M.R. and plasma cholesterol levels were also measured.

The cases were classified as hyper-, eu- or hypothyroid on the evidence presented by the clinical findings at the time of the test and during a follow-up period which varied from 6 to 21 months. It should be emphasized that in many cases the diagnosis could not be made with absolute certainty, but the group in which the patient was placed seemed on balance, after a very careful examination of the evidence, most accurately to describe the state of his or her thyroid function. Two cases had to be discarded, one due to inadequate follow-up, the other because no decision could be made even after considering all the available evidence. A few illustrative case histories are given later.

A number of patients received two or more tests during the period under review. If there was no significant difference in the results or change in the clinical findings, and if no form of treatment had taken place between the tests, the results were averaged and treated as one test. If, however, any of the above three criteria was not complied with, the separate tests were included in the series. Finally, we were left for our analysis with 110 tests made on 99 different patients. Their distribution between the three groups of thyroid function is shown in Table I. Each group is subdivided into those patients who had received no previous treatment for thyroid disorder, and those who had been previously treated by either surgery, iodine 131 or drugs. Among the last mentioned class none had been taking thiouracil, neomercazole or thyroid extract, for at least 6 weeks prior to the test, except two hypothyroid patients who had been off thyroid for one and four weeks beforehand respectively. Some cases had received more than one form of treatment, and have been classified under the heading corresponding to the most recent treatment received before the test.

**Table I**

*Distribution of tests*

Total number of tests 110; of patients 99; figures in brackets indicate number of patients where different from number of tests.

| Thyroid function | Total number | No previous treatment | Previous treatment | | | |
|---|---|---|---|---|---|---|
| | | | Surgery | $I^{131}$ | Drugs | Total |
| Hyperthyroid ..... | 42 (41) | 27 | 4 | 3 | 8 (a) | 15 (14) |
| Euthyroid ..... | 55 (50) | 30 (28) | 4 | 15 (12) | 6 (b) | 25 (22) |
| Hypothyroid ..... | 13 | 5 | 2 | 2 | 4 | 8 |

a) 5-thiouracil, 2-neo-mercazole, 1-thyroid extract; b) 4-thiouracil, 1-thyroid extract, 1-Lugol's iodine.

1 patient hypothyroid and 4 patients euthyroid after treatment with $I^{131}$ had tests done before therapy and therefore also occur in hyperthyroid group.

## Results

The maximum % uptakes and corresponding 48 hr protein bound iodine concentrations in plasma are shown for all the tests in the scatter diagram (Diagram 2). We will first confine our attention to the hyper- and euthyroid patients. It was found by plotting cumulative frequency curves on probability paper that the values of maximum % uptake and the log plasma iodine concentration (either total or protein bound fraction) approximated to normal distributions about their mean values in the different groups.

The number of wrong diagnoses has sometimes been used as an index for comparing the reliability of different tests. FRYERS (1956) has criticised this practice on the grounds that it places undue emphasis on a small number of cases and neglects the remainder. In assessing his results he assumed that cases belonged to either a euthyroid or hyperthyroid popu-

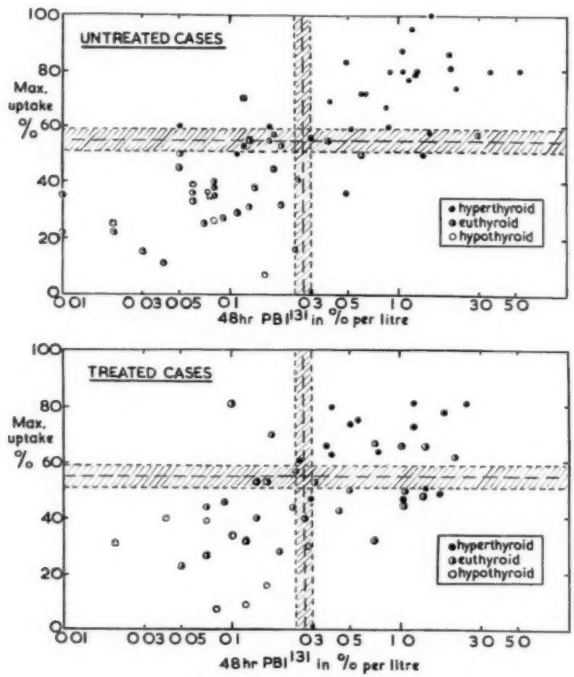


Diagram 2. Scatter diagram showing results of maximum uptake and 48 hr $PBI^{131}$ tests. The shaded bands indicate the chosen upper limits of normal with the estimated possible errors at these levels.

lation each having a normal statistical distribution, and then proceeded to calculate the standard deviation of the difference between the two means as a percentage of the difference between the means. This percentage was then used as an index of reliability. Although this method does make use of all the data and is a valid means of comparison, the figure of merit obtained does not immediately help the physician using the test to know the percentage of correct diagnoses he may expect. We have therefore used an index which may be termed 'the percentage overlap' for comparing the reliability of our tests. If the results of a test performed on hyper- and euthyroid patients are normally distributed about two mean values, then there is an intermediate value such that the percentage of the results in the hyper- and euthyroid groups lying respectively below and above this value are equal. Suppose the means of the two groups with their standard deviations are $x_1$ and $\sigma_1$, $x_2$ and $\sigma_2$, respectively, then the intermediate value, $\bar{x}$ such that the percentage of the upper group having values below $\bar{x}$ equals the percentage of the lower group having values above $x$ is given by the equations:

$$\bar{x} - x_1 = A \cdot \sigma_1 \text{ and } x_2 - \bar{x} = A \cdot \sigma_2 \text{ where } A \text{ is a constant.}$$

Hence eliminating $\bar{x}$ we have $A = \dfrac{x_2 - x_1}{\sigma_1 + \sigma_2}$ and from tables of the normal distribution the probability of values occurring outside a range of $x \pm A\sigma$ may be found. The required 'percentage overlap' is half of this probability, and shows immediately the probable number of wrong diagnoses that may be expected. In Table II the results of our three tests are compared in this way.

**Table II**

*Summary of results*

| Test | Hypothyroid | | Euthyroid | | | | Hyperthyroid | | | | % overlap between hyper- and euthyroid groups | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Untreated | | Treated | | Untreated | | Treated | | | |
| | Mean | $\sigma$ | Mean | $\sigma$ | Mean | $\sigma$ | Mean | $\sigma$ | Mean | $\sigma$ | Untreated | Treated |
| Maximum % uptake | 26 | 12.2 | 39 | 14.6 | 49 | 14.4 | 72 | 14.8 | 66 | 12.7 | 13 | 27 |
| log plasma concentration (%/l at 48 hrs) × 100 | 1.12 | 0.35 | 1.29 | 0.39 | 1.57 | 0.49 | 1.99 | 0.45 | 1.94 | 0.32 | 20 | 32 |
| log protein-bound iodine conc. (%/l at 48 hrs.) × 100 | 0.88 | 0.33 | 1.01 | 0.50 | 1.48 | 0.47 | 1.91 | 0.44 | 1.88 | 0.33 | 17 | 31 |
| Effective half-life (days) | 5.1 | 2.42 | 6.2 | 1.32 | 5.3 | 1.64 | 4.7 | 1.40 | 4.7 | 1.54 | 29 | 43 |

*Untreated cases.* It is convenient first to discuss the results obtained in those patients who had received no previous treatment for thyroid disorder.

GOODWIN et coll. (1951) concluded from their series that measurement of protein-bound activity at 48 hours was a superior test to the measurement either of total plasma activity or of maximum uptake in the gland. This conclusion was based on the numbers of patients wrongly classified by each test. FRYERS (1956) however, considered that their results did not demonstrate that measurement of plasma activity was more specific than that of protein-bound activity. He therefore only included measurements of whole blood activity in his studies, and concluded that this was a much inferior test to maximum uptake. Our results confirm this finding, but also show that the measurement of protein-bound activity is rather better than that of total plasma activity, though it is still not so good as that of maximum uptake. Also the difference in reliability between the uptake and total plasma measurements does not appear to be so great in our series as was found by FRYERS.

It is possible that some of the conflicting conclusions reached by different workers about the superiority of one or other radio-iodine diagnostic test are caused by the different compositions of their euthyroid and hyperthyroid groups.

Poorer correlation between results of radio-iodine uptake tests and the clinical findings in patients with non-toxic or toxic nodular goitres as compared with normal euthyroid patients or those with primary thyrotoxicosis have been reported by a number of authors (WERNER et coll. (1949), KEATING et coll. (1950), MILLER et coll. (1951), FREEDBERG and URELES (1950), HORST (1954 and 1955), MCCONAHEY et coll. (1956).

**Table III**

*Results on untreated patients grouped according to presence or absence of goitre in euthyroids and type of hyperthyroidism*

| | Euthyroid | | | | Hyperthyroid | | | | % overlap between euthyroids without goitre and primary thyrotoxic group |
|---|---|---|---|---|---|---|---|---|---|
| | No goitre | | Goitre present | | Primary thyrotoxicosis | | Toxic nodular | | |
| No. of tests (no. of patients in brackets) | 16 (15) | | 14 (13) | | 19 (19) | | 8 (8) | | |
| | Mean | $\sigma$ | Mean | $\sigma$ | Mean | $\sigma$ | Mean | $\sigma$ | |
| Maximum % uptake | 39 | 13.3 | 39 | 16.4 | 75 | 12.2 | 64 | 18.2 | 8 |
| log protein-bound iodine conc. (%/l at 48 hrs.) $\times$ 100 | 0.91 | 0.41 | 1.13 | 0.58 | 2.01 | 0.41 | 1.66 | 0.48 | 9 |

HORST (1955) also found that using the 48 hour protein-bound activity as an index, a few cases with non-toxic goitre gave values above the upper limit of normal. The number of non-toxic and toxic nodular goitres in a series is therefore likely to influence comparisons of different tests. This is well brought out by our results.

Table III shows the values obtained in the untreated euthyroid and hyperthyroid groups further subdivided into euthyroid with and without a goitre present, and primary thyrotoxicosis and toxic nodular goitre. The two euthyroid groups have the same mean value of maximum uptake, though those with goitre present have a greater dispersion about the mean (as shown by the values of $\sigma$). The mean protein-bound activity is slightly higher for the non-toxic goitres. The values obtained by both tests are lower for toxic nodular goitres than for cases of primary thyrotoxicosis. It should be pointed out that the differences between the means of the subdivisions are not statistically significant at the 5 % level of probability. Contrary to the findings of GOODWIN et coll. (1951) the protein-bound activities for the two hyperthyroid groups have the greatest difference and when tested by the t-test give $t = 1.94$, $n = 25$, $P = 0.065$. The last column of Table III indicates that if all our hyperthyroid patients had been in the primary thyrotoxic group and none of our euthyroids had had any goitre present, the efficiency of our tests would have been greater and we should have found very little difference in reliability between maximum uptake and protein-bound activity measurements.

*Previously treated cases.* Examination of Table II shows that in the diagnosis of patients having had some form of previous treatment, all tests are much less reliable. The maximum % uptake and the total plasma activity are still respectively the most and least efficient of the three tests. The mean values for the patients hyperthyroid after treatment are all slightly less than those for the untreated hyperthyroid group, but the differences are not statistically significant. The composition of the treated group is similar to that of the untreated group as regards the proportion of toxic nodular goitres (5 out of 15 as compared with 8 out of 27). The increased mean values in the patients euthyroid after some form of treatment is much more marked, and the differences are statistically significant. It is of interest to examine this group in more detail and Table IV shows the results further sub-divided according to the nature of the previous treatment. Of the four patients having had previous surgery, two showed values of both uptake and 48 hour protein-bound activity above normal (see later discussion for our choice of normal limits). One of these had been operated on for a toxic nodular goitre 13 years previously and had exophthalmos, but was otherwise euthyroid. The other had been operated on for primary thyrotoxicosis 17 years previously and at the time of our test had a metastasis in the neck from a carcinoma of the ovary.

**Table IV**

*Mean values of results obtained on euthyroid previously treated patients when grouped according to the nature of the previous treatment*

| | Previous treatment | | |
|---|---|---|---|
| | Surgery | $I^{131}$ | Drugs |
| Maximum % uptake ..... | 55 | 47 | 46 |
| log protein-bound iodine conc. (%/l at 48 hrs.) × 100..... | 1.41 | 1.66 | 1.09 |

*Case 1.* G. N., a married woman, who had a right thyroidectomy in 1935 and a left thyroidectomy in 1938. At both times she was suffering from thyrotoxicosis.

In 1954, at the age of 55 years, she had a carcinoma of the ovary which was incompletely removed. The operation was followed by a course of deep roentgen therapy. She remained well for 9 months following the treatment, but then complained of tiredness, palpitations, sweating and loss of weight. She said she felt she was becoming thyrotoxic again. At the time she appeared anxious and had a 4 × 4 cm fixed mass in her left neck behind the sternomastoid. Her skin was warm and moist. There were no eye signs, tremor or thyroid enlargement.

Her pulse was reg. 90; and B. P. 160/80 mm Hg. Plasma cholesterol: 163 mg/100 ml. B.M.R.: Readings varied from + 17 to + 26 %, but the estimation was considered unsatisfactory. Radio-iodine test: Maximum uptake 66 %; 48 hr plasma activity 1.17 %/litre; 48 hr $PBI^{131}$ 1.01 %/litre. An isocount map of the neck showed no appreciable uptake in the lump.

The patient was not clinically toxic, and the mass in her neck was regarded as a metastatic gland from her ovarian carcinoma. She gradually developed other metastases and died a year later. During the year she showed no signs of thyrotoxicosis and the thyroid histology at post-mortem showed areas of non-toxic colloid adenomatous change with marked fibrosis on the left.

The other two patients in this group had been operated on for primary thyrotoxicosis ($2^1/_2$ years previously) and a non-toxic goitre (9 years previously), respectively, and showed normal values of both uptake and protein-bound activity.

Silver et coll. (1955) reported one case euthyroid after surgery who had a high uptake and 72 hr $PBI^{131}$. Raised protein-bound or plasma activities associated with normal uptakes in patients hypo- or euthyroid after surgery have been reported by Ansell et coll. (1951), Blom and Terpstra (1953) and Barry and Pugh (1953). In this connection the following case from our hyperthyroid group is of interest.

*Case 2.* A. F., a married woman of 51 years, who in July 1955 attended hospital with severe thyrotoxicosis and exophthalmos. She was treated with neo-mercazole and improved clinically, but later developed agranulo-cytopaenia. Neo-mercazole was stopped and she recovered from the agranulo-cytopaenia. She was then given Lugol's iodine and a partial thyroidectomy was performed in September 1955. Two months later she was

referred for radio-iodine studies, as she was thought to have developed recurrent thyrotoxicosis. At the time she was extremely agitated, never stopped talking, and had the appearance of thyrotoxicosis, with bilateral exophthalmos and a tremor. She had no sweating. Her pulse was reg. 120, and B.P. 180/110 mm Hg. Radio-iodine test: Maximum uptake 47 %; 48 hr plasma activity 1.16 %/litre; 48 hr $PBI^{131}$ 1.04 %/litre. A therapeutic dose of 6 mC of radio-iodine was prescribed, calculated to give 7,000 rep to a thyroid estimated clinically to weigh 20 g. 48 hours after the administration of the therapeutic dose, an isocount map of the neck was made to investigate the possibility of retrosternal extension. None was revealed, but the comparison of the map with one done on an 18 g model thyroid indicated that the weight of the patient's thyroid might have been as low as 8 g. If this was so, then 18,000 rep were delivered.

In March 1956 the patient felt better, but showed signs of myxoedema. She was gaining weight and felt the cold; the skin of her arm was very dry and scaly. Plasma cholesterol was 500 mg/100 ml. She was put on thyroid by mouth which she has continued to take. In October 1956 she was well and leading a normal life, although still nervous and rather agitated. There were no signs of myxoedema, but exophthalmos was still present.

The fact that the patient developed myxoedema is not surprising in view of the high therapeutic dose that was probably given. Clinically the patient was hyperthyroid at the time of the test, so that in this case the $PBI^{131}$ reflected the clinical state more accurately than the maximum uptake.

In the group of 6 patients euthyroid after treatment with some form of drug, all had protein-bound activities within the normal range (note mean value of log protein-bound iodine concentration in Table IV), but two patients of the four previously treated with thiouracil had high uptakes (79 % and 70 %, 10 months and 26 months after the end of treatment, respectively). The case history of one of these was as follows.

*Case 3.* L. K., a single woman of 66 years, who, following a period of physical and mental strain, was admitted to hospital in 1952 with left ventricular failure, hypertension and coronary thrombosis. After a month, she was discharged on thiouracil and digitalis, but as her follow-up visits to hospital distressed her, she did not keep up attendance.

She stayed on thiouracil for two years, and during the last 3 months of this period, her thyroid had greatly enlarged. This enlargement decreased when thiouracil was stopped in January 1954. A radio-iodine test was performed in May 1954. During the last year she had been losing weight, and her hair had been falling. She felt all extremes of temperatures, and her appetite was fair. Her past history was of rheumatic fever at 15 years, a nervous breakdown at 48 years, and a high blood pressure since that age. On examination, the patient appeared to be a nervous woman with a fine tremor of her hands, thin dry hair and dry skin. She had no abnormal eye signs. Her thyroid was nodular and estimated by palpation to weigh between 50 and 60 g. Her heart was enlarged to the left, but there was no evidence of cardiac failure. Her pulse was reg. 90, and B.P. 200/120 mm Hg. Plasma cholesterol: 400 mg/100 ml. B.M.R. + 10 %.

Radio-iodine test: Maximum uptake 85 %; 48 hr plasma activity 0.2 %/litre. The clinical picture was of hypertension and controlled auricular fibrillation, with her thyroid function at the lower limit of normal. It was felt advisable to leave her in this condition because of her old cardiac infarct.

The radio-iodine test was repeated 3 months later with the following result: Maximum uptake 92 %; 48 hr plasma activity 0.03 %/litre. The result of a test done in October 1954, after another 3 months, was: Maximum uptake 79 %; 48 hr plasma activity 0.13 %/litre;

48 hr $PBI^{131}$ 0.11 %/litre. A year later a further test gave almost the same values, *i. e.*: Maximum uptake 83 %; 48 hr plasma activity 0.11 %/litre; 48 hr $PBI^{131}$ 0.08 %/litre.

Since there had been no treatment or clinical change between these last two tests, their results were averaged and counted as one test in our series. (The first two tests were not included as no measurements were made of $PBI^{131}$.)

The patient's clinical state from September 1954 to October 1955 was judged to be euthyroid. In November 1955 she developed severe cardiac pain and a therapeutic dose of radioiodine was given in an attempt to make her myxoedematous, and to relieve her cardiac condition. As a result of this treatment she felt better and was able to get about more with less dyspnoea. During the 9 months after treatment, her weight remained stationary with her pulse at reg. 100 and her B.P. at 200/100 mm Hg.

The other two thiouracil treated patients had uptakes of 44 and 40 %, 36 and 40 months after the end of treatment, respectively. Again, SILVER et coll. (1955) report a raised uptake in a patient non-goitrous after stopping thiouracil therapy.

Raised uptakes in patients euthyroid after treatment with $I^{131}$ have been noticed by several authors (FREEDBERG and URELES 1950, HORST 1954, BENUA and DOBYNS 1955, SILVER et coll. 1955. The raised mean value shown in Table IV for this group does not necessarily confirm these findings. Of the 3 tests that gave values above the normal limits, two were on a patient who had been treated with thiouracil up to 16 months before the $I^{131}$ therapy, and the third was on a patient who had been treated by surgery 17 years previously. The mean of eight tests where $I^{131}$ had been the only form of treatment was 41 %, *i. e.* not significantly different from that of the untreated euthyroid group.

The increased mean value of log protein-bound activity after $I^{131}$ therapy is however a genuine effect and confirms the findings of ANSELL et coll. (1951), HORST and KUHLENCORDT (1954), LARSSON (1955) and SILVER et coll. (1955). The mean of the eight tests where only $I^{131}$ therapy had been given was 1.47. The time elapsed between treatment and tests in the whole group of patients euthyroid after $I^{131}$ therapy varied from 2 to 29 months with a mean of 12 months. Nine of the fifteen tests showed protein-bound activities above normal limits, but there seemed to be no correlation between the occurrence of these higher values and the time after treatment. It was noted, however, that all nine tests were on patients suffering originally from primary thyrotoxicosis, while 3 of the 6 tests showing normal values were on patients who had originally had toxic nodular goitre, and as mentioned previously these patients may have had lower values of $PBI^{131}$ before treatment. The data were not available to verify this point.

Our conclusions are therefore that in patients euthyroid after treatment raised values of maximum % uptake may occur after surgery or thiouracil, while raised protein-bound activities may be found after surgery or $I^{131}$ therapy. These increased values may persist for periods of years.

*Upper limits of normal.* For day to day interpretation of radio-iodine diagnostic tests it is necessary to select some upper level of normal above which results may be taken as a strong indication of thyrotoxicosis. From what has been said already such an upper limit chosen in order to produce the least number of diagnostic errors might need to be different for treated and untreated groups of patients. However, it seems a more practicable procedure to choose a level which will give the least number of diagnostic errors on untreated patients, and then use this level with caution on treated patients, taking into consideration from the patient's past history where errors are likely to occur.

From the scatter diagram (Diagram 2) 55 % appears to be a suitable upper limit of normal for maximum percentage uptake, and is comparable with the value used by a number of other workers (HORST 1954, NEWBURGER et coll. 1955).

For 48 hour protein-bound activities MACGREGOR et coll. (1953) took 0.2 % and 0.4 % per litre as the upper limit in normals and the lower limit in hyperthyroids, respectively. For patients with intermediate values a conversion ratio (*i. e.* PBI/total plasma activity) greater than 0.5 was taken to indicate thyrotoxicosis. Our experience does not confirm this. In our cases with protein-bound activities between 0.2 and 0.4 % per litre, conversion ratios varied from 0.6 to 1.0 although only 7 out of the 15 tests were on patients clinically hyperthyroid. The work of BARBIERI (1955) supports this finding in that at 48 hours he found that the mean value of the conversion ratio for euthyroid patients was 0.82. We have therefore chosen a definite value of 0.27 % per litre as the upper limit of normal 48 hour protein-bound activity. This level is not very different from 0.25 % per litre used by HORST (1954). For total plasma activity 0.5 % per litre is the upper limit of normal that gives the least number of wrong diagnoses in our untreated patients.

Table V shows the number of wrong diagnoses using each of the three tests. The figures bear out the general conclusions reached by the statistical treatment given earlier. For untreated cases the numbers of wrong diagnoses given by the uptake and protein-bound activity tests are equal, but in fact two of the uptake errors (57 % for two euthyroid cases) are not significantly greater than the chosen upper limit, when one considers the accuracy of the measurements. The percentage failures are slightly lower than the calculated percentage overlap, and although this might not be apparent in a larger series, it could conceivably be caused by genuine deviations from the statistically normal distribution of the tests in one or more of the groups.

All except one of the untreated cases wrongly diagnosed by the uptake test were toxic nodular or non-toxic goitres. Of those wrongly diagnosed by the measurement of protein-bound activity, three were toxic nodular

**Table V**

*Wrong diagnoses in hyper- and euthyroid groups*

| Test | No. of clinically euthyroid patients diagnosed as hyperthyroid by test | | No. of clinically hyperthyroid patients diagnosed as euthyroid by test | | Total No. of wrong diagnoses |
|---|---|---|---|---|---|
| | Untreated | Treated | Untreated | Treated | |
| Maximum % uptake ..... | 3 | 7 | 3 | 4 | 17 |
| Plasma activity at 48 hours . | 4 | 9 | 5 | 4 | 22 |
| Protein-bound activity at 48 hours ..... | 3 | 11 | 3 | 1 | 18 |
| Maximum % uptake and 48 hrs. protein-bound activity combined ..... | 1 | 4 | 5 | 5 | 15 |

or non-toxic goitres, and one was a patient with Hashimoto's disease who gave a value of 0.7 % per litre at 48 hours, although having no clinical signs of toxicity. It is interesting to note that GOODWIN et coll. (1951) also found a high value of protein-bound activity in a similar case associated with normal uptake, and values of blood cholesterol and BMR on the hypo- side of the normal range. The other two failures of the protein-bound activity test were a euthyroid patient having a Parkinsonian type of tremor (0.38 % per litre), and a case of mild thyrotoxicosis which was misdiagnosed on both tests (50 % maximum uptake and 0.11 % per litre) and subsequently successfully treated with $I^{131}$.

The failures in cases euthyroid after some form of treatment have already been discussed. Of the four tests which gave normal uptake in cases thyrotoxic after some form of treatment, two were on the same patient; the first gave 49 % 8 months after treatment with neo-mercazole, the second 51 % 5 months after radio-iodine therapy. The remaining two were 47 % in a patient 2 months after surgery, and an equal value in a case with toxic nodular goitre, who had been treated with thyroid tablets for myxoedematous symptoms about a year previously. It will be noted that all these uptake values might be classified as 'high' normal. The one treated hyperthyroid case diagnosed as normal by the protein-bound activity test was a patient previously treated with thiouracil. The result was 0.25 % per litre which in view of the accuracy of the measurements can hardly be considered as significantly below the upper limit of normal.

Some improvement in diagnostic accuracy may be obtained by using the uptake and $PBI^{131}$ test in conjunction. If we diagnose as toxic only those patients in whom both tests give values above normal, then as shown in the bottom line of Table V the errors are reduced to 15 (*i. e.*

15.5 % of the total number of tests on hyper- and euthyroid patients). This improvement is, however, gained entirely in the treated group of patients, and the tendency to diagnose as euthyroid patients who are in fact hyperthyroid is increased, particularly in the untreated group.

*Hypothyroid patients.* Our tests have been disappointingly unsuccessful in distinguishing patients with myxoedema. The group was too small to subdivide in any way and there was no obvious difference between results on treated and untreated cases. The mean values of uptake, plasma and protein-bound activities are also shown in Table II, and although these are all lower than the euthyroid group the differences are not sufficient to ensure any high degree of accuracy in diagnosis. The smallest percentage overlap between the hypothyroid and untreated euthyroid group is 33 obtained by the uptake test. The results reported by BARBIERI (1955) suggest that the conversion ratio at 48 hours might be a reasonably successful method for the diagnosis of myxoedema. This is not confirmed by our results, the mean conversion ratio of our untreated euthyroids and hypothyroids being 67 and 62 % respectively with a percentage overlap between the two groups of 47.

For maximum percentage uptake a lower limit of normal of 20 % gives three euthyroid patients diagnosed as hypothyroid, but 9 of the 13 hypothyroid patients have uptakes within the normal range. The myxoedematous cases reported in most published series have been few but have usually had uptakes below 15 % (WERNER et coll. 1949, KEATING et coll. 1950). GOODWIN et coll. (1951) report one case with an uptake of 23 % while THODE et coll. (1953) found five cases with uptakes over 20 %, the highest being 29 %. Our highest uptake (40 %) was obtained in a patient who had been off thyroid for a period of six weeks prior to the test, and may have been due to some residual effect since PALEY et coll. (1955) have found that high $PBI^{131}$ and clearance rates may occur at least one month after thyroid administration has been stopped. A value of 39 % occurred in a patient with acromegaly who had had thyroid up to a year previously. Another high value (34 %) was in a patient who developed myxoedematous symptoms 43 years after thyroidectomy, and may be related to the high uptakes found in some of our cases euthyroid after surgery. The highest protein-bound activity (0.28 % per litre) was also found, associated with a 30 % uptake, in a patient hypothyroid 16 years after surgery, while the two patients hypothyroid after $I^{131}$ had uptakes below 20 % but protein-bound activities of 0.12 and 0.16 % per litre. However, although previous treatment may affect the results in hypothyroid patients in a similar manner to that discussed earlier for euthyroid patients, it must be pointed out that only one of our untreated hypothyroids had an uptake of less than 20 %, and this was a congenital case with an associated protein-bound activity of 0.16 % per litre. Of

the remaining four untreated hypothyroids an uptake of 39 % was found in a case concurrently suffering with diabetes mellitus, 35 % in a patient successfully treated for carcinoma of the breast three years previously, 26 % in a patient also having osteo-arthritis, and 25 % in a patient having no other complications. The case history of the patient with the 39 % uptake was as follows.

*Case 4.* B. Q., a single woman of 49 years, who attended as an outpatient in November 1955 with a three months history of excessive thirst, lassitude, and polyuria. During the past year, her hair and eyebrows had been coming out, and she had suffered with occipito-frontal headaches. She felt the cold badly, and for 6 months her eyelids and face had been swollen and puffy. She had gained some two stone (ten kg) in weight over the past few years. There was slight dyspnoea on exertion and some ankle swelling. Her blood sugar was 0.28 %; pulse reg. 88; B.P. 200/100 mm Hg.

A provisional diagnosis of hypertension, myxoedema and diabetes mellitus was made. The latter condition responded to diet, and a month later the patient was referred for radio-iodine studies. At this time her diabetes was controlled by diet. The tests were done in mid-December, and repeated at the end of January 1956. On both occasions similar results were obtained, namely maximum uptake 39 %; 48 hour plasma activity 0.09 and 0.05 %/litre, respectively; 48 hour $PBI^{131}$ 0.09 and 0.03 %/litre respectively. The results of these two tests were averaged and counted as one in our series.

In February 1956 the patient started to take thyroid by mouth, and her condition improved. She lost about 5 kilos in weight over 6 months, became much more active, and her hair and eyebrows became thicker. In September 1956 she was still on thyroid, and was working and living a normal life.

We are forced to the conclusion that except insofar as a low uptake (*i. e.* below 20 %) may confirm hypothyroidism our tests by themselves can contribute very little help to the physician where the clinical diagnosis is in doubt. However, used in conjunction with thyroid stimulating hormone, they may help to distinguish between primary myxoedema and hypopituitarism (QUERIDO and STANBURY 1950; PERLOFF et coll. 1951). The following case demonstrates such use, even though the result of the first radio-iodine test was in the normal range.

*Case 5.* A. C., a single man of 55 years, who had a 9 year history of headaches, a 2 year history of lethargy and a slowing of his mind with a tendency to fall asleep. He had also had a loss of axillary and pubic hair, and of libido. For a year he had been taking 3 grains of thyroid daily. His B.M.R. was — 11 %; plasma cholesterol 328 mg/100 ml. He presented a clinical picture of hypopituitarism, causing secondary myxoedema, and radio-iodine tests were undertaken as part of a full investigation.

The results of the first test, done in June 1955, one week after he had stopped taking thyroid, were: Maximum uptake 31 %; 48 hr plasma activity 0.04 %/litre; 48 hr $PBI^{131}$ 0.02 %/litre.

The patient was then given thyroid stimulating hormone, I.M., 5 units t.d.s. for one day, and 10 units q.d.s. for 7 more days. The radio-iodine test was then repeated, and gave the following result: Maximum uptake 65 %; 48 hr plasma activity 1.93 %/litre; 48 hr $PBI^{131}$ 1.87 %/litre.

The result of the first test was included in our series as a hypothyroid case, but the normal value of uptake may well have been due to the patient having so recently been on thyroid. The increased values after T.S.H. helped to confirm that the endocrine failure was primarily pituitary.

Subsequent treatment with cortisone, testosterone and thyroid considerably improved the patient's condition. Thirteen months later, he died in hospital from a carcinoma of the bronchus with metastases. Post-mortem confirmed the diagnosis of Simmonds' disease.

*Half-life in gland.* No clear evidence of a double exponential decay was found, and the values quoted below are based on the half-lives of the single exponential curves that best fitted the measured points. It was not expected that the half-life would provide any useful diagnostic information and this in fact proved to be so. The mean values in the different groups are also shown in Table II. The mean for the untreated euthyroids is significantly higher than that for all the other groups except the hypothyroid, *i. e.* hyperthyroidism is associated with a more rapid decay from the gland which persists even after successful treatment. This is in agreement with the findings of HORST and KUHLENCORDT (1954) and LARSSON (1955).

It is again of some interest to study the treated euthyroid group in more detail. In this group 11 tests gave 48 hr $PBI^{131}$ activity greater than 0.27 % per litre. The mean half-life of these tests (4.1 days, $\sigma = 1.32$) was significantly lower than that of the remaining fourteen tests whose mean was 6.2 days ($\sigma = 1.29$) *i. e.* the same as that of the untreated euthyroid group. LARSSON (1955) divided his treated euthyroid group at the 0.5 % per litre level, and although the mean half-life for those with 48 hr $PBI^{131}$ below 0.5 % per litre was the longer of the two, both means were lower than that for untreated euthyroids. It is possible that if the division had been made at 0.27 % per litre his results would have been similar to ours, namely that normal levels of $PBI^{131}$ were associated with normal half-lives even after treatment. The inference is that a contributing cause of the high values of 48 hour $PBI^{131}$ found in patients euthyroid after treatment is the continued more rapid rate of decay from the gland. This factor will not however affect the maximum uptake, which is therefore likely to be a more reliable index of the return of clinical euthyroidism after treatment.

## Discussion

It is not to be expected that any test will accurately distinguish between the three groups of thyroid function, since in fact there can be no clear-cut dividing lines. A correct diagnosis is one which leads to a correct treatment of the patient, and it is as a guide to treatment in cases where the clinical picture is not clear, that a laboratory test for thyroid

function should be able to provide useful information. One must therefore be able to place a high level of confidence in its results. In our experience, the radio-iodine tests discussed above still leave much to be desired from this standpoint, since under the best conditions misleading results are likely to be obtained in about 20 % of all cases.

The reasons for these errors may be inherent in the very nature of the radio-iodine tests, since they do not measure the absolute amount of either the stable iodide trapped by the thyroid, or the circulating thyroid hormone. Uptake results will therefore be influenced by the size of the total miscible iodide pool in the body. SILVER et coll. (1955) discuss this point more fully, and FRASER (1956) has shown how a combination of drug-loading with the radio-iodine test may help to remove anomalous results in cases of iodine deficiency. Tests measuring plasma activity or $PBI^{131}$ will be affected by the size of the stored hormonal iodine pool, as has been pointed out by SILVER et coll. (1955) and LARSSON (1955). The latter found normal levels of serum protein-bound iodide in patients euthyroid after treatment, even when the same patients gave values of 48 hour $PBI^{131}$ above normal. It is in this group that the determination of serum protein-bound iodide may, in spite of its greater technical difficulty, prove to be of the most value.

However, the comparatively simpler radio-iodine tests have a usefulness that is enhanced by an awareness of the most common errors from which they suffer. There also seems to be some advantage in carrying out more than one type of radio-iodine test, since they do not all necessarily produce misleading results in the same cases, and experience can teach us upon which to place most reliance in a particular case. Such experience can only be gained by careful comparison of results with clinical diagnoses. In many such comparisons that have been published, perhaps too much emphasis has been placed on the successes rather than the failures. If we have erred in the opposite direction, it is because we feel that an acute sense of the limitations of a laboratory test is essential if the maximum value is to be gained from its use.

## Conclusions

Analysis of the results and comparison with the clinical findings lead to the following conclusions.

For distinguishing between hyper- and euthyroid patients, the maximum percentage uptake gives slightly better results than the 48 hour protein-bound activity, but the difference is small and there is probably little to choose between the two tests. Both are superior to the measurement of 48 hour total plasma activity.

Results on untreated cases are better than those on cases having had some form of treatment for thyroid disorder.

In untreated cases diagnostic errors are more likely to occur in cases of toxic nodular or non-toxic goitre.

In previously treated cases the most common fault is for the test to indicate hyper-thyroidism when the patient is in fact euthyroid. One may find raised uptakes after thiouracil, raised 48 hour $PBI^{131}$ after $I^{131}$ therapy, and raised values of both after surgery. Some improvement in diagnosis may be obtained in previously treated cases by only considering the simultaneous occurrence of increased uptake and increased 48 hour $PBI^{131}$ as evidence of hyper-thyroidism.

Upper limits of normal may be taken as 55 % maximum uptake in the gland, total activity in plasma at 48 hours 0.5 % per litre, and protein-bound activity at 48 hours 0.27 % per litre of plasma.

All the tests (including the conversion ratio at 48 hours) give little help in distinguishing between hypo- and euthyroid patients, except in so far as an uptake below 20 % is strong evidence of myxoedema, but a large proportion of myxoedematous patients have uptakes above this value.

The effective half-life in the gland has little diagnostic value. It is generally shorter in hyperthyroidism and cases euthyroid after treatment. In the latter group, shorter half-lives tend to be associated with raised values of 48 hour $PBI^{131}$.

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## SUMMARY

Measurements of maximum percentage uptake of radio-iodine into the thyroid gland, the 48 hour plasma activity, and the 48 hour $PBI^{131}$ have been made in 99 patients. The value and shortcomings of the test as shown by comparison with the clinical findings in certain types of cases are discussed.

## ZUSAMMENFASSUNG

Messungen der maximalen prozentuellen Aufnahme von radioaktivem $I^{131}$ in der Schilddrüse, der 48 Stundenaktivität in Plasma und der nach 48 Stunden vorhandenen Menge von proteingebundenem $I^{131}$ sind an 99 Patienten gemacht worden.

Der Wert und die Nachteile des Testes im Vergleich zu den klinischen Befunden bei gewissen Typen von Fällen werden besprochen.

## RÉSUMÉ

Les auteurs ont mesuré chez 99 malades le pourcentage maximum de fixation de l'iode radio-actif dans la thyroïde, l'activité du plasma à la 48e heure et le PBI$^{131}$ (I$^{131}$ protéinique) à la 48e heure. Par comparaison avec les résultats de l'examen clinique ils étudient l'intérêt et les imperfections de ce test dans certains types de cas.

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# EARLY EFFECTS OF WHOLE BODY IRRADIATION ON CHOLINESTERASE ACTIVITY IN GUINEA-PIGS' BLOOD, WITH SPECIAL REGARD TO RADIATION SICKNESS

by

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The increasing use of atomic energy and radioactive isotopes in research and industry as well as the risks of accidental irradiation and radioactive contamination in atomic warfare have greatly augmented the interest in the biologic and medical effects of ionizing radiations and the search for simple but reliable methods for an early detection of radiation injury has become urgent. The symptoms of radiation sickness may not appear until days or even weeks after the irradiation (DUNHAM et coll. 1951, inter alios), but for the appropriate handling and treatment of the victims, especially in an atomic disaster, the possibility of estimating the severity of the exposure as early as possible, even before subjective symptoms appear, is desirable. For this reason the changes which occur and the diagnosis of radiation injury in the early stages are of great interest.

In addition to the reduction in the number of white and red blood corpuscles, other morphologic and biochemical changes are present before the subjective symptoms. Thus, early degenerative and mitotic changes have been found in the bone marrow (BLOOM 1948, THORELL 1955, inter alios). CASTER and ARMSTRONG (1955) have reported changes

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in the electrolyte balance in the blood and HAYES and HEWITT (1955) and MILCH and co-workers (1956) in the serum lipo-proteins. BERNSTEIN et coll. (1956) have found an approximately twofold increase in the total plasma amino acid concentrations in chicks at one hour following irradiation and a threefold increase after three hours. A similar alteration in plasma amino acids has been reported in man by KATZ and HASTERLIK (1955). Numerous other alterations in the blood and urine chemistry have also been found (PROSSER 1950, inter alios), although these are stated to be mostly late changes.

A number of investigations have been devoted to the effects of irradiation on various enzyme systems especially the phosphatases and phosphorylases (PETERSEN et coll. 1953, VAN BEKKUM 1955, EICHEL 1955, MOLLURA and GOLDFEDER 1956, FEINSTEIN 1956), as well as the desoxyribonucleases (DOUGLASS and DAY 1955). There is usually an increase in the phosphatase activity in the spleen but the findings are inconsistent, and the increase is interpreted as due to a decrease in the organ weight (EICHEL 1955, FEINSTEIN 1956).

The cholinesterases are also among the enzyme systems which have been studied following irradiation. Despite the great interest in this enzyme system at the present time, there are relatively few reports on the effects of irradiation in vivo on cholinesterase to be found in the literature.

LÜTHY (1953) found in roentgen irradiated rats a decrease in the activity of cholinesterase in the serum after 500 r and 1,000 r although, following 250 r, the values usually lay within the normal limits. After 500 r, that is a little less than the $LD_{50}$ for rats, there was often a decrease in enzyme activity which was manifested as early as a few hours after the irradiation. The results were, however, inconsistent, some animals the more the lower the dose given, showing an increased activity. The same variable results were obtained in guinea-pigs by ORD and STOCKEN (1953). After 1,000 r, that is 4 to 5 times the $LD_{50}$, they generally observed a drop in the unspecific cholinesterase activity 6 hours after irradiation, with a tendency to a recovery in 48 hours. They found no changes in the unspecific or specific cholinesterase in the intestinal muscles of guinea-pigs 24 hours after 1,000 r.

In mice exposed to 25 to 300 r total body roentgen irradiation SABINE (1956) noticed an increase in the cholinesterase activity of the erythrocytes 4 days after the exposure. High values were observed after 300 r on the third to fifth days. After that there was a sharp decrease and values far below the normal were usual at the end of the first week; recovery was complete after three weeks.

BURN, KORDIK and MOLE (1952) could demonstrate a 50 per cent drop in the intestinal unspecific cholinesterase activity in the rat 48

hours after irradiation, a slight decrease being apparent as early as after 24 hours. No significant changes were found in the specific cholinesterase activity. CONARD (1952, 1956) reported a significant decrease in the cholinesterase activity in the small intestine of rats 15 to 20 hours after irradiation with recovery after three weeks. No correlation of the enzyme activity changes with intestinal motility was found.

In vitro experiments show no significant changes in the cholinesterase activity after roentgen irradiation (KWIATKOWSKI 1938) at least not after doses less than 5,000 r (LÜTHY 1953). The latter obtained only a 7.5 per cent inhibition after 10,000 r, but for higher doses there was a linear relation between the irradiation dose and the inhibition of the serum cholinesterase. SABINE (1956) found that the activity of whole human plasma cholinesterase was unaffected by 100,000 r with 1,000 $kV_p$ roentgen rays. Purified electric eel cholinesterase exposed in dilute sheep serum to 250 $kV_p$ roentgen rays was inactivated at a rate of 15 per cent per 100,000 r.

The scope of the present investigation was to study the early changes in the cholinesterase activity in the blood of guinea-pigs exposed to relatively high doses of roentgen irradiation. It seems particularly desirable to study the possibility of using a simple routine method for the determination of cholinesterase activity developed in our institute, for the early diagnosis of radiation injury. In one series the cholinesterase activity was determined in whole blood sampled on filter paper and in another the activities were determined in the erythrocytes and plasma separately. In a few animals the cholinesterase activity was studied in the liver and the bone marrow.

## Material and Methods

*Experimental animals.* About one hundred male guinea-pigs were used; the average body weight was 420 $\pm$ 100 g.

*Irradiation data.* 260 kV, 10 mA, filter 4 mm Al + 0.4 mm Cu. F. T. D. 70 cm and 40 cm respectively, in the two series described below.

The animals were kept in plastic boxes during the exposure. The results to be described are based on animals exposed to 400 r. The $LD_{50/14}$ for guinea-pigs is 175 to 250 r (SPECTOR 1956), and the dose given was chosen to obtain a definite action. The dose rates were 38 r/min. and 123 r/min., respectively.

*Blood sampling.* For the determination of the esterase activity in the whole blood the method described by HEILBRONN (1953) was employed. Drops of blood from an ear vein were collected on filter paper (Munktell No. 8). After drying, blood spot disks 1 cm in diam. were punched from the paper, two such disks being used for each single determination. The quantity of blood taken each time from an animal was about 0.5 ml. In this series blood could be collected more than once after the exposure without rendering the animals anaemic. This method is called "the blood spot method" in this paper.

For the determination of the esterase activity in the red corpuscles and plasma separately, for which a somewhat larger quantity of blood was needed, the blood was

**Table 1**

*Experimental conditions in determination of cholinesterase activity*

| *Blood spot method* | *Separated blood method* |
|---|---|
| *Source of enzyme*<br>Two blood-soaked and dried circular spots of filter paper (Munktell No. 8) 1 cm diameter, each divided into 3 parts. | *Source of enzyme*<br>3 ml of either<br>a suspension of hemolyzed red cells or<br>a solution of plasma in water.<br>Red cell suspension:<br>The centrifuged blood sample was relieved of plasma and an equal volume of aq. redest. added. 1.4 ml of this suspension was diluted to 25 ml with aq. redest.<br>Plasma solution:<br>1.4 ml of the plasma removed from blood sample after centrifuging was diluted to 25 ml with aq. redest. |
| *Buffer used*<br>3 ml of a modified Michel's buffer<br>Composition:<br>4.123 g (0.02 mol) sodium barbital<br>0.5446 g (0.04 mol) $KH_2PO_4$<br>44.730 g (0.6 mol) KCl<br>28 ml of 0.100 mol HCl<br>aq. redest. to 4,000 ml | *Buffer used*<br>3 ml of a modified Michel's buffer<br>Composition:<br>The same as for blood spot method but made up with aq. redest. to 2,000 ml only. |
| *Substrates:*<br>0.3 ml of a solution of acetyl-$\beta$-methylcholine iodide (MeCh) giving a concentration in the reaction mixture (3.3 ml) of $1.84 \times 10^{-2}$ mol $l^{-1}$, or<br>0.3 ml of a solution of butyrylcholiniodide (BuCh) giving a concentration in the reaction mixture (3.3 ml) of $2.01 \times 10^{-2}$ mol $l^{-1}$. | *Substrates:*<br>0.6 ml of MeCh or BuCh giving the same final concentrations in the reaction mixture (6.6 ml) respectively, as in blood spot method. |

collected in a heparinized centrifuge tube by heart puncture. The blood was centrifuged and the plasma was carefully removed and substituted by an equal amount of distilled water. In order to make the procedure rapid, the erythrocytes were not washed.

The mean blood sample volume taken with this method, which in this paper is called "the separated blood method", was $2.6 \pm 0.5$ ml. The mean blood volume of guinea-pigs is 7.2 ml per 100 g body weight (Ancill 1956). According to this, the quantity of blood taken from each animal in the present investigation was calculated to be $9.1 \pm 2.4$ per cent of its total blood volume. In this series blood was taken only once before and once after the irradiation. Thus, for the determination of the enzyme activity at various inter-

vals (one and two days) after the irradiation, different animals had to be used in order to avoid too large a blood loss. The controls were treated equally excepting in not being exposed to irradiation.

*Blood cell volume and number.* In most cases the packed red cell volume was determined in a hematocrit centrifuge (International Hemacrit Centrifuge) with heparinized capillary tubes according to the method of GUEST and SILER (1934). The volume of the white blood cells has not been taken into account. The number of red cells was counted in a Bürker counting chamber.

*Cholinesterase determination.* The two cholinesterases in the blood, specific and unspecific, were studied. Specific cholinesterase, called AChE (acetylcholinesterase) according to AUGUSTINSSON and NACHMANSOHN (1949) in this paper, is present in guinea-pigs both in the erythrocytes ($AChE_E$) and plasma ($AChE_P$). (See *e. g.* ZAJICEK and DATTA 1953.) Unspecific cholinesterase, called ChE (cholinesterase) (AUGUSTINSSON and NACHMANSOHN 1949) in this paper, is present only in plasma ($ChE_P$).

For the determination of the enzyme activities, the method described by TAMMELIN et coll. (1951, 1952, 1953) was used. The substrates employed were acetyl-$\beta$-methylcholine iodide (MeCh) and butyrylcholine iodide (BuCh). (For the concentrations used see Table 1.)

The activities of $AChE_E$ and $AChE_P$ were determined with MeCh and the activity of ChE with BuCh. Thus, in the blood spot method the activity determined with MeCh represents both the $AChE_E$ and $AChE_P$ activities. In the separated blood method it is possible to determine the activities of the enzymes individually. Though the erythrocytes were not washed, the possible influence of $AChE_P$ in the erythrocyte fraction was considered negligible.

The buffers used were modifications (TAMMELIN 1953, HEILBRONN 1953) of those described by MICHEL. (See Table 1.)

*Bone marrow and liver.* Bone marrow was taken from the hind legs 24 hours after irradiation. The marrow was weighed and the AChE and ChE activities were determined with MeCh and BuCh as substrates. These activity values, referred to unit weight of marrow, were compared with those of the non-irradiated controls.

Four g of the finely cut liver were weighed and homogenized in a buffer solution. An equal amount was dried to constant weight. The enzyme activities were determined in the same way as in the bone marrow. They were referred to unit weight of dry liver and compared with the activities obtained in the non irradiated controls.

For the statistical treatment Student's t-test was used.

## Results

The results are based on enzyme activity determinations in 102 guinea-pigs: 15 with the blood spot method, 49 with the separated blood method, and 15 for activity in the bone marrow and 23 in the liver. The irradiated animals died within 8 to 14 days.

*Blood spot method.* The results with the blood spot method have earlier been briefly presented (LUNDIN, CLEMEDSON and NELSON 1955). In general, there was a distinct increase of the AChE activity and a decrease of the ChE activity in the blood during the first two days after the irradiation as compared with the initial enzyme activities, *i. e.* the activities obtained in the same animals before the irradiation (Diagram 1).

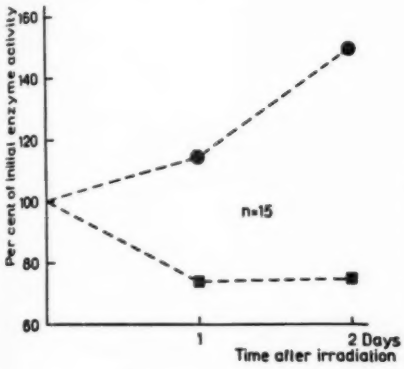


Diagram 1. Changes in acetylcholinesterase activity of red blood corpuscles ($AChE_E$) and plasma ($AChE_P$) ——●—— and the cholinesterase activity of plasma ($ChE_P$) ——■—— after whole body roentgen irradiation with 400 r. Each point represents a mean value of 15 (n) animals. (Blood spot method.)

These results seemed to be promising as regards the aim of this investigation, but as it was obvious that the blood spot method was in certain respects unsatisfactory, we tried to confirm them with the more accurate separated blood method.

*Separated blood method.* As the packed red cell volume differs considerably from one animal to another it is necessary with this method to correct the $AChE_E$ activity by means of determinations of this volume. As is seen in Diagram 2 and Table 2 the red cell volume changes in the same way in the irradiated and the control animals. It had decreased to about 80 per cent of the initial value as early as the first day and remained lowered during the following day. The difference is statistically significant when comparing the initial values with those of the second blood sampling. This is valid for the irradiated as well as the control animals, with the exception of those of the control animals on the second day when the difference is only probable. The decrease of the red cell volume is probably an effect of the blood sampling. Such an effect, which could not be ascribed merely to the quantity of blood removed, has earlier been described in dogs by HANNA and MARSHALL (1955). The same also applies to the number of red cells (Table 2 and Diagram 3) although the significance is less. For neither the volume nor the number

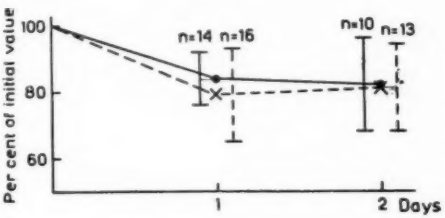


Diagram 2. Changes in volume of red blood corpuscles. ——●—— control animal. — —×— — animals irradiated with 400 r. Vertical lines show standard deviations, and n is the number of animals in each point. (Separated blood method.)

**Table 2**

*Results of statistical treatment*

Symbols for P as expressed below:

| | | |
|---|---|---|
| — | $(P > 0.05)$: | indicates the difference as not probable |
| * | $(0.05 > P > 0.01)$: | » » » » probable |
| ** | $(0.01 > P > 0.001)$: | » » » » highly probable |
| *** | $(0.001 > P)$: | » » » » statistically significant. |

| Probability (P) of | Irradiated | | Controls | |
|---|---|---|---|---|
| | 1st day | 2nd day | 1st day | 2nd day |
| | Comparing initial with 2nd sample | | | |
| Changes in red cell volume... | *** | *** | *** | *** |
| Changes in number of red cells ..... | ** | * | ** | * |
| | Comparing volume with number of red cells | | | |
| Changes in percentage ..... | — | — | — | — |
| | Comparing $AChE_E$ with $AChE_P$ | | | |
| Changes in percentage ..... | * | *** | — | — |
| | Comparing $AChE_E$ with $ChE_P$ | | | |
| Changes in percentage ..... | *** | *** | * | * |
| | Comparing $AChE_P$ with $ChE_P$ | | | |
| Changes in percentage ..... | * | — | — | — |

of red cells do the changes from the initial value show any significant differences except when comparing the changes in the number of red cells on the first day (see Table 3). Although these differences exist, there is no clear evidence of any significant alterations in the relationship between the red cell volume and the number of red cells.

Diagrams 4 to 6 show the changes in the activities of $AChE_E$, $AChE_P$ and $ChE_P$ in the irradiated and control animals. The $AChE_E$ activity (Diagram 4) increases by about 20 per cent in both groups of animals already on the first day and remains high during the second day. The standard deviations, however, are comparatively large.

The $AChE_P$ activity of the irradiated animals decreases to about 75 per cent of the initial value on the second day. In the non-irradiated controls on the other hand, the $AChE_P$ activity remains unchanged during the first two days (Diagram 5); the standard deviations are, however, again rather large.

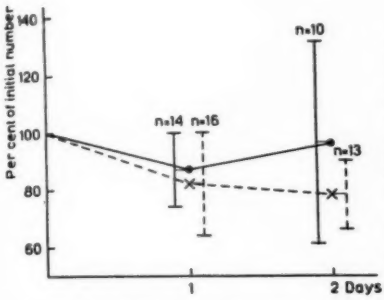


Diagram 3. Changes in number of red blood corpuscles in guinea-pigs. For symbols see diagram 2.

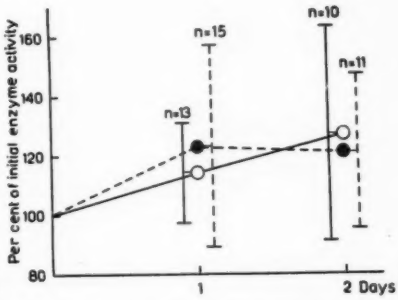


Diagram 4. Changes in acetylcholinesterase activity of red blood corpuscles ($AChE_E$) in guinea-pigs. ——○—— control animals. — —●— — animals irradiated with 400 r. Vertical lines show standard deviations; n is the number of animals in each point. (Separated blood method.)

Diagram 6 shows that the $ChE_p$ activity decreases to about 70 per cent during the first day and then remains at that level during the following day. In the control group the activity is unchanged during the first day but there is a tendency to somewhat lower activity values on the second day. The standard deviations of the $ChE_p$ activity values are smaller than those of the $AChE_E$ and $AChE_p$. One possible explanation of this is the fact that the numerical values of the $ChE_p$ activity were higher throughout than those of the two other enzymes.

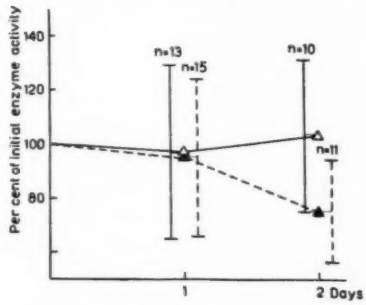


Diagram 5. Changes in acetylcholinesterase activity of plasma ($ACHE_p$) in guinea-pigs. ——△—— control animals. — —▲— — animals irradiated with 400 r. For further description see legend of diagram 4.

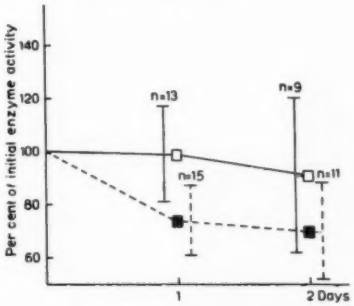


Diagram 6. Changes in cholinesterase activity of plasma ($ChE_p$) in guinea-pigs. ——□—— control animals. — —■— — animals irradiated with 400 r. For further description see legend of diagram 4.

**Table 3**

*Results of statistical treatment*

| Probability (P) of differences in | Comparing irradiated with controls 1 day | 2 days | 1 day with 2 days irradiated | controls |
|---|---|---|---|---|
| Changes in red cell volume ..... | — | — | — | — |
| Changes in number of red cell ..... | ** | — | — | — |
| Changes in $AChE_E$ activity..... | — | — | — | — |
| Changes in $AChE_p$ activity..... | — | * | * | — |
| Changes in $ChE_p$ activity ..... | *** | — | — | — |

For explanation of symbols see table 2.

The results of the statistical treatment of the data are presented in Tables 2 and 3. It is evident that a statistically significant difference exists between the irradiated and control animals only as far as the $ChE_p$ activity is concerned. The difference in $AChE_p$ activities between these two groups on the second day is probable (Table 3). The change in $AChE_p$ activity in the irradiated animals from the first to the second day is also probable. Three animals have been omitted from the material before the statistical treatment owing to obvious errors during the experimental procedure.

In Table 2 the results of a mutual comparison of the changes in the activities of the various enzymes are shown. It is seen that the differences are more pronounced for the irradiated than for the control animals, that comparing the changes in the activity of $AChE_E$ with those of $ChE_p$, these differences are statistically significant (irradiated animals) or probable (controls), and that the changes in the activity of $AChE_p$ seem to follow those of $ChE_p$.

Thus, the statistical treatment of the values essentially confirms the conclusions which can be drawn from the curves.

Using both the blood spot and the separated blood method, we have followed the changes in enzyme activity in some animals for several days. However, the number of animals being small in these series, and as the standard deviations are fairly large, we do not feel justified to draw any definite conclusions from that material at present. Furthermore, this would be beyond the scope of this investigation, which deals only with the early changes after irradiation. The experiments with determination of the enzyme activities in the liver and bone marrow have only been of a preliminary nature but further investigations are going on.

## Discussion

The occurrence and activity of cholinesterases in the blood differ considerably in different species (ZAJICEK and DATTA 1953), a fact that must be taken into consideration when studying the effects of various agents on the enzyme activity. In dogs and guinea-pigs the activities per unit volume are sufficiently high to enable accurate determinations. This fact and the desirability to be able to expose several animals to irradiation at the same time suggested the use of guinea-pigs in the present investigation. Furthermore, the experimental animals must be large enough to tolerate repeated small losses of blood (blood spot method) or the loss of a comparatively large initial blood volume (separated blood method).

This investigation has shown that the roentgen irradiation of guinea-pigs lowers the cholinesterase activity in the plasma. This effect which is especially pronounced for the $ChE_p$ activity, persists during the first two days after irradiation which is in accordance with the results obtained by ORD and STOCKEN (1953) in guinea-pigs and LÜTHY (1953) in mice. Several authors report a lowering of the activity of ChE in other tissues as well as in the small intestine of the guinea-pig (BURN et coll. 1952, CONARD 1956).

Thus, the results from several investigations seem to indicate that ChE is sensitive to irradiation. The real nature of this sensitivity is still unknown but at least three principle possibilities may be briefly mentioned.

1) *The synthesis of ChE may be damaged by the irradiation.* It is generally supposed that $ChE_p$ is synthesized chiefly in the liver. Though no definite proof of this seems to have been presented, many investigations have been performed concerning the correlation between $ChE_p$ activity and liver diseases. Several authors (*e. g.* BURN et coll. 1952, DOULL and CUMMINGS 1954) have assumed or actually found a lowering of the ChE in the liver after irradiation. So far, however, nobody seems to have ascribed this lowered activity exclusively to an impaired synthesis. In radiation injury there is an abnormal break-down of proteins (ORD and STOCKEN 1953), and KATZ and HASTERLIK (1955) and BERNSTEIN et coll. (1956) have reported a considerable increase of plasma amino acids some hours after irradiation. This, however, does not necessarily mean a decomposition of the $ChE_p$.

2) *The enzyme molecule may be damaged by the irradiation.* If so, it is likely that plasma irradiated in vitro should also display a decreased ChE activity. Only very high doses, however, have been found to have such an effect (KWIATKOWSKY 1938, LÜTHY 1953, SABINE 1956).

3) *The irradiation may cause the formation of abnormal metabolites which are cholinesterase inhibitors.* This might be the subject of far-reaching speculations but there are still too few facts known to permit of a fruitful discussion.

The AChE activity in plasma seems to be affected by irradiation to the same extent as the ChE activity. As the reason why AChE is present in rather large amounts in the guinea-pig plasma is still unknown, as is the relation between AChE and ChE in plasma, it would be in vain to comment any further upon this finding.

The AChE activity in erythrocytes was shown to increase during the two first days after irradiation. This result was obtained both with the blood spot method and the separated blood method. A similar increase was, however, found with the separated blood method also in the non-irradiated controls. The increased $AChE_E$ activity, therefore, may be ascribed to the blood sampling rather than to the irradiation, and it has not been possible to demonstrate any significant change in the $AChE_E$ activity in this investigation. This is in agreement with the findings of DOULL and CUMMINGS (1954) in rats. SABINE (1956) on the other hand was able to show an appreciable increase in the $AChE_E$ activity in mice after roentgen irradiation with doses in excess of 25 r. This increase was not manifested until the 4th day after irradiation and therefore his results are not inconsistent with our findings.

The object of this investigation was to find out if there are any early changes in the cholinesterase activities in blood and, if so, whether the simple routine method for cholinesterase determinations (blood spot method) adopted in our laboratory could be used as an early diagnostic tool of irradiation injury.

However, the actual conditions in this respect are unfavourable, depending especially on the large variations both in the normal cholinesterase activities and in the changes of these activities after irradiation. Nevertheless our results, the two methods confirming each other, as well as those obtained by other authors, seem to justify further research along similar lines.

**Acknowledgement**

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## SUMMARY

Male guinea-pigs suffered a loss of cholinesterase activity in plasma amounting to about 30 per cent during the first two days after whole body roentgen irradiation with 400 r. No changes in the activity of acetylcholinesterase in erythrocytes to be ascribed to the irradiation could be demonstrated during the first two days. A routine method for determination of the cholinesterase activities has been tested but is found less suitable as a diagnostic tool for the early recognition of the effects of irradiation.

## ZUSAMMENFASSUNG

Nach Ganzkörperbestrahlungen mit Dosen von 400 r erlitten männliche Meerschweinchen innerhalb der beiden ersten Tage einen Verlust der Cholinesteraseaktivität im Plasma von ungefähr 30 %. Keine auf die Bestrahlung zu beziehende Veränderung der Aktivität der Acetylcholinesterase in den Erythrocyten konnte während der ersten beiden Tage demonstriert werden. Eine Rutinmethode zur Bestimmung der Cholinesteraseaktivitäten ist durchgeprüft worden; sie wurde jedoch als diagnostisches Mittel für die frühzeitige Erkennung von Bestrahlungseffekten weniger geeignet befunden.

## RÉSUMÉ

Des cobayes mâles ont présenté, pendant les deux premiers jours après une irradiation de tout le corps par 400 r, une baisse de l'activité cholinestérasique du plasma d'environ 30 pour cent. Il n'a pas été possible de mettre en évidence, pendant les deux premiers jours, de modification de l'activité de l'acétylcholinestérase des érythrocytes imputable à l'irradiation. Les auteurs ont essayé une méthode de routine pour mesurer l'activité cholinestérasique, mais l'ont trouvée peu utilisable comme procédé de diagnostic précoce des effets de l'irradiation.

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# A SIMPLIFIED LIGHT-TRAP

by

*Morris Owen*

The design of light-traps to dark-rooms has undergone little change for many years. Currently recommended structures include open mazes, mazes with curtains or doors, simple two-door traps, and revolving doors. While all of these may be made completely light-tight, none has attained the simplicity of ingress or egress which is necessary for the convenient operation of a dark-room. From the point of view of convenience in use, the ideal light-trap should have no turns, no doors or curtains, and indeed, occupy no floor space. This ideal is of course impossible. A light-trap may, however, be made which is without obstruction and virtually without turns. The premise on which the design is based is that a roentgen dark-room need not be completely unilluminated. An object of dark-room illumination is in effect to provide as much light as possible for the normal handling time plus an adequate safety margin without fogging films.

A light-trap designed on this premise has been found satisfactory from the viewpoints of convenience, light exclusion, and floor space occupied. The design and dimensions are shown in the figures. The position and strength of the lights in the 'light' part of the processing suite is important, but there is no difficulty in arranging adequate illumination.

The ceiling and three of the four walls of the light-trap are lined with slats. (The slats are assembled parallel to one another much like a Venetian blind.) It will be seen that each pair of slats acts as a miniature light-trap; any light which may be reflected will be so reduced by subsequent 'trappings' that the usefulness of the dark-room is unimpaired. The angles

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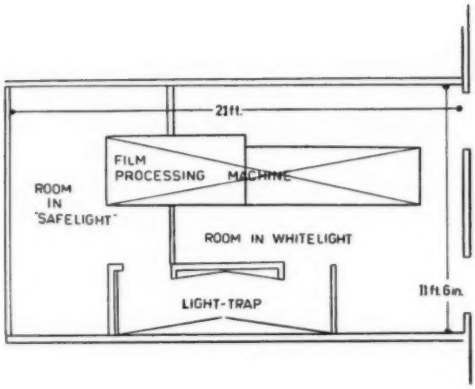


Fig. 1. Plan of processing suite showing position of light-trap in relation to 'safe-light' and 'white-light' subdivision.

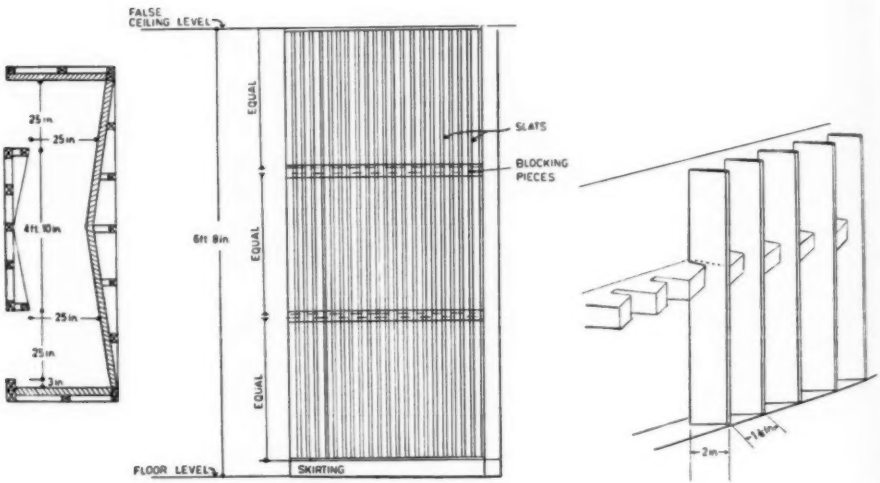


Fig. 2. a) Measurements of light-trap. The hatching indicates the walls lined with slats. b) Elevation of slatted wall with measurements. c) Detail of arrangement of slats with supporting spacer.

at which the slats are placed would ideally vary, and be formed by the radii of a circle with its centre outside the light-trap entrance; for convenience of construction a mean angle was prescribed and all the slats angled similarly. The finish is in matt black.

This light-trap has been in use in a suite with mechanical processing handling up to three hundred films per day. Little maintenance has been found necessary; in six years the only attention necessary has been the replacement of a few broken slats, and repainting.

**Acknowledgement**

I wish to thank STEPHENSON and TURNER, architects to the hospital, for permission to publish the blue-prints and drawings. The reproductions are by Janette McDonald, The Royal Newcastle Hospital.

## SUMMARY

A dark-room light-trap suitable for use in a roentgen department, is described. Its features are the small area occupied, ease of ingress and egress, and the freedom from obstructions.

## ZUSAMMENFASSUNG

Eine Dunkelzimmerschleuse, die sich zum Gebrauch in einer Röntgenabteilung eignet, wird beschrieben. Ihre charakteristischen Kennzeichen sind die kleinen Ausmasse, das einfache Hinein- und Herausgehen sowie das Fehlen jeglicher Sperre.

## RÉSUMÉ

Description d'un 'piège à lumière' pour l'accès du laboratoire de développement d'un service de radiologie. Il se caractérise par la petite surface occupée, par la facilité d'entrée et de sortie et par l'absence d'obstacles.

FROM THE INSTITUTE OF RADIOPHYSICS (DIRECTOR: PROF. R. SIEVERT) AND RADIUMHEMMET (DIRECTOR: PROF. S. HULTBERG), KAROLINSKA SJUKHUSET, STOCKHOLM, AND THE RADIOPHYSICS LABORATORY (DIRECTOR: S. BENNER), KING GUSTAF V JUBILEE CLINIC, GOTHENBURG, SWEDEN

# MEASUREMENTS OF RELATIVE BETA DEPTH DOSES IN 'TISSUE EQUIVALENT' MATERIAL

by

*Gunnar Walinder*

The aim of the investigation was to devise measuring methods for determination of beta depth doses in wet tissue from semi-infinite isotope solutions. The measurements were made with photographic film and a scintillation probe specially designed for the purpose.

The following isotopes were investigated:

**Table 1**

| *Substance* | $^{198}$Au | $^{32}$P | $^{76}$As |
|---|---|---|---|
| Mean energy ($\beta$) ..... | 0.32 MeV | 0.70 MeV | 1.17 MeV |
| $k_\gamma$ ..... | 2.2 | — | 2.0 |
| Max. range in water .. | 3.8 mm | 8.0 mm | 15.7 mm |

In the above table, $k_\gamma$ is the dose rate in rad/hour which a punctate gamma source with an activity of 1 mC gives at a distance of 1 cm in water.

*Theory.* When beta particles from radioactive isotopes (usually with energies below about 3 MeV) impinge upon an absorbant medium, the principal effects are as follows:

1. Inelastic collisions with the electrons of the absorbant medium, giving rise to ionizations and being the principal cause of the energy losses.
2. Elastic collisions with the atomic nuclei of the absorber, which give rise to scattering of the incident electrons without losses of energy.

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The loss of energy per length unit can be expressed by the well-known Bethe-Bloch formula:

$$-\frac{dE}{dx} = \frac{2\pi NZe^4 \varrho}{A \cdot mv^2}\left\{\ln \frac{mv^2 E}{2I^2(1-\beta^2)} - (2\sqrt{1-\beta^2} - 1 + \beta^2)\ln 2 + \right.$$

$$\left. + 1 - \beta^2 + \tfrac{1}{8}(1-\sqrt{1-\beta^2})^2\right\} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (1)$$

where

| | | |
|---|---|---|
| $N$ = Avogadros number | $Z$ = Atomic number | $\varrho$ = Density of absorber |
| $\beta$ = v/c | $m$ = Electron mass | $E$ = Kinetic energy of electron |
| $c$ = Velocity of light | $e$ = Electron charge | $I$ = Potential of mean excitation |
| $v$ = Velocity of electron | $A$ = Atomic weight of absorber | |

Elastic scattering of the electrons is caused chiefly by interaction with the atomic nuclei of the absorber. In defining the scattering it is customary to take four groups, depending on the number of collisions, namely: (1) Singular scattering (scattering caused by a single collision); (2) Plural scattering (from two to about twenty collisions); (3) Multiple scattering (here the scattering has a Gaussian distribution, and the range comprises a number of collisions from about twenty up to an angular distribution having a mean angle of 20 degrees); and lastly (4) diffuse scattering, *i. e.*, when a state of equilibrium has occurred and the angular distribution changes from Gaussian to a $\cos^2$-distribution (4, 7, 8, 20).

In order to gain some idea of the variation of angular distribution with absorber depth, we shall deduce an approximate formula for the mean angle. The following description is largely based on deductions of ROSSI & GREISEN (21) and BETHE & ASHKIN (4).

In respect of singular scattering, RUTHERFORD's formula for minor directional changes of the incident particles may be employed (since the cross-section decreases very swiftly with increasing angular deviation) on the following assumptions:

1. The nuclear charge is assumed to give rise to a purely Coulombian field; *i. e.*, the screening caused by the atomic electrons as well as the finite extension of the nucleus, are disregarded.
2. The absorbant material is assumed to consist of atoms having a low $Z$-value, *i. e.*, the Born approximation can be applied (this approximation presupposes that $Z/137\,\beta \ll 1$).
3. The electron spin is disregarded.

For the present investigations, conditions 2 and 3 are satisfactorily fulfilled but, at very small scattering angles, corrections are required because of the screening.

Let us now assume that the probability of an electron with momentum $p$ and velocity $\beta c$ being subjected, while traversing distance $dx$, to collisions which deflect its direction through an angle $\Theta$ from the original direction into the solid angle $d\omega$, is $P(\Theta)\,d\omega dx$, where

$$P(\Theta)\,d\omega\,dx = \frac{q_R N}{A}\,d\omega\,dx = \frac{4NZ^2 r_e^2}{A}\left(\frac{mc}{\beta p}\right)^2 \frac{d\omega}{\Theta^4}\,dx \quad \ldots\ldots\ldots\ldots\ldots\ldots (2)$$

where $q_R$ is RUTHERFORD's cross-section, $dx$ is the absorber depth in g/cm², and $r_e = e^2/mc^2$ is the classical electron radius.

For the square of mean angle $(\Theta)^2_{mean}$ after a short distance $x$ g/cm² ($x$ must be sufficiently small for no appreciable mean energy loss to have occurred; and usually it is so for the range where the scattering is multiple (See 7)): we obtain, if $d\omega = 2\pi\Theta d\Theta$,

$$(\Theta)^2_{\text{mean}} = \int_0^x \int_{\Theta_{\min}}^{\Theta_{\max}} \Theta^2 \, P(\Theta) \, 2\pi \, \Theta d\,\Theta dx = \int_0^x \int_{\Theta_{\min}}^{\Theta_{\max}} \frac{8\pi NZ^2 r_e^2 \left(\frac{mc}{\beta p}\right)^2 d\Theta dx}{\Theta} =$$

$$= \frac{8\pi NZ^2 r_e^2}{A}\left(\frac{mc}{\beta p}\right)^2 x \ln \frac{\Theta_{\max}}{\Theta_{\min}} \quad \ldots\ldots\ldots\

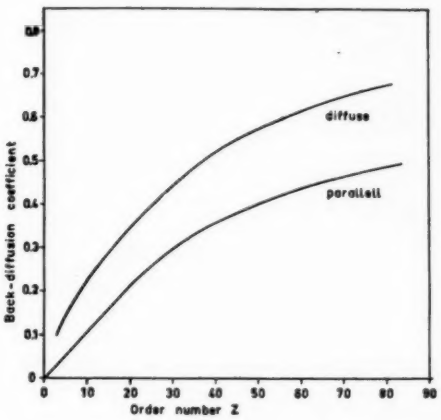


Diagram 1. Back-scattering coefficient (at saturation) for normal incidence, and for diffuse angular distribution of the incident beam. After PAUL & STEINWEDEL.

In the above diagram the back-scattering coefficient is given as a function of $Z$. (The back-scattering coefficient is the number of back-scattered electrons divided by the number of incident primary electrons at saturation.) The curve is based on values obtained from different sources, and will be found in (20). In accordance with BOTHE's theory (6), the back-scattering coefficient is independent of the energy of the incident electrons.

Taking equation (1) and (4) as a basis, we shall now try to define the conditions required if a substance is to be considered — from the standpoint of beta-radiation — equivalent to water. For this purpose, water is assumed to be equivalent to wet tissue.

The energy loss per unit of length will, according to eq. (1) be proportional to $Z/A$ if the depth is measured in g/cm² (*i. e.*, $\varrho = 1$) and if the logarithmic expression in (1) is assumed to be constant — a plausible assumption if the comparison relates solely to substances with reasonably similar $Z$ values, and hence similar $I$ values. As an analogue, the scattering as in equation (4) will be proportional to $Z/\sqrt{A}$ or, if we also take into account the previously disregarded scattering due to interaction with the absorber's electrons, $\sqrt{Z(Z+1)/A}$. If two substances are to be comparable in the radiation sense, then their $Z/A$- and $\sqrt{Z(Z+1)/A}$-values have to be in agreement. This is naturally impossible where elements are concerned but in the case of chemical compounds and mixtures of different substances the composition may be so chosen as to give effective values for $Z/A$ and $\sqrt{Z(Z+1)/A}$ that will accord with the corresponding values for the substance it is desired to replace (14).

We now define:

$$\left(\frac{Z}{A}\right)_{eff} = \sum_n \frac{A_n}{M} \cdot \frac{Z_n}{A_n} = \frac{\sum_n Z_n}{M}$$

and similarly for the scattering

$$\left(\sqrt{\frac{Z(Z+1)}{A}}\right)_{eff} = \sqrt{\sum_n \frac{A_n}{M} \cdot \frac{Z_n(Z_n+1)}{A_n}} = \sqrt{\frac{\sum_n Z_n(Z_n+1)}{M}}$$

For mixtures in which the fraction of the weight for substance 1 is $a_1$, for substance 2, $a_2$, and so on, of the total weight of the mixture, we obtain, respectively,

$$\left(\frac{Z}{A}\right)_{eff} = \sum_i a_i \frac{\sum_n Z_{in}}{M_i}$$

and

$$\left(\sqrt{\frac{Z(Z+1)}{A}}\right)_{eff} = \sqrt{\sum_i a_i \frac{\sum_n Z_{in}(Z_{in}+1)}{M_i}}$$

where $\sum_n$ is the sum of the atoms in the respective molecules, and $\sum_i$ is the summation of the molecules in the mixture.

The effective values of a few different substances are tabulated below.

**Table 2**

| Substance | $(Z/A)_{eff}$ | $(\sqrt{Z(Z+1)/A})_{eff}$ | $\varrho$ g/cm³ | Chem. formula |
|---|---|---|---|---|
| Water | 0.555 | 2.05 | 1.0 | $H_2O$ |
| Mix D | 0.556 | 1.90 | 0.99 | See below! |
| Gelatin | 0.512 | 1.96 | | $C_{102}H_{51}N_{31}O_{39}$ |
| Triacetate | 0.528 | 1.97 | 1.36 | $n[C_6H_7O_5(COCH_3)_3]$ |
| Scint. cryst. | 0.538 | 1.84 | 1.05 | n(CH) |
| Lucite | 0.540 | 1.92 | 1.18 | $n\left[\begin{array}{l} CH_3 \\ -CCH_2- \\ COOCH_3 \end{array}\right]$ |

Mix D is a mixture prepared by JONES and RAINE (13), consisting of 60.8 % paraffin wax ($C_{25}H_{52}$), 30.4 % polyethene n(CH), 6.4 % MgO, and 2.4 % $TiO_2$.

*Experimental.* A scintillation probe was designed for the purposes of these measurements.

In measuring beta depth doses with scintillation crystals, an endeavour has been made to reduce the thickness of the crystals in order to secure more accurate values for the dose. The literature contains reports of dosimeters where antracene crystals with a thickness of 0.1 mm have been used. With these, however, it is difficult to avoid light guides between crystal and photomultiplier. In the measuring of isotopes having beta energies similar to those investigated by the present author, a very appreciable fluorescence and Cerenkov radiation will arise in the light guide and seriously interfere with the measuring results (1, 2, 3, 23). The scintillation probe described is based on a different principle.

Fig. 1 shows the principles of the mode of procedure. A crystal 2 cm in length (*i. e.*, longer than the maximum beta range), consisting of p-terphenyl plus tetraphenylbutadiene in polystyrene (see Table 2) is inserted in Mix D, which in turn is surrounded by water.

In these measurements the photomultiplier current was recorded after gradual shifts of the crystal (0.1 mm at a time), and each such value was subtracted from the preceding one.

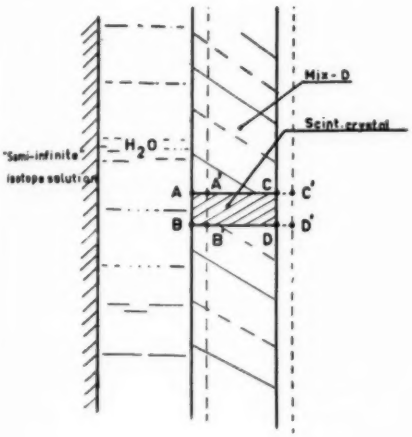


Fig. 1. Mode of action of the scintillation probe. The positions of the scintillation crystal before and after a shift (ABCD and A'B'C'D' resp.). The photomultiplier is situated at CD and C'D', resp.

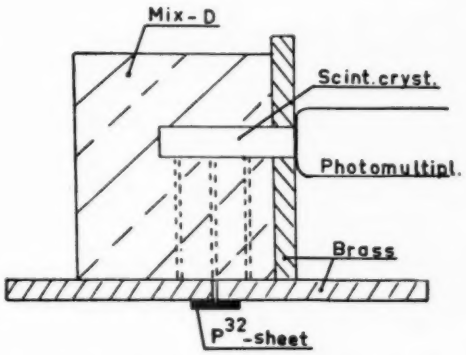


Fig. 2. Equipment for control of the reflection ability of Mix D.

In Fig. 1 the position of the crystal is shown before and after one shift (ABCD and A'B'C'D'). On the assumption of adequate reflection at the lateral surfaces of the crystal — so that the effect of the altered solid angles under which the scintillations are 'seen' by the photomultiplier when the position is shifted from ABCD to A'B'C'D' will be negligible — the difference between the scintillation effects in the two positions will be equal to the original effect in AA'BB'. When the shifts are as small as 0.1 mm the relevant difference is assumed to be proportional to the dose in AA'BB'.

Mix D contains MgO (giving the mixture its white colour), which is an excellent reflector of those light wave-lengths that arise in the crystal.

The reflection ability of Mix D was controlled as follows.

The crystal was surrounded by Mix D, in which three small holes had been drilled. The brass diaphragm (the horizontal brass plate at the base in Fig. 2) was progressively moved so that its hole coincided, in turn, with each of the holes in the phantom. Since no difference in the photomultiplier current was detectable, the reflecting power of Mix D was evidently satisfactory.

Fig. 3 shows the design of the scintillation probe. The isotope solution is placed in lucite containers that can be raised or lowered by screws. Some of these containers were cylindrical ('semi-infinite' sources of

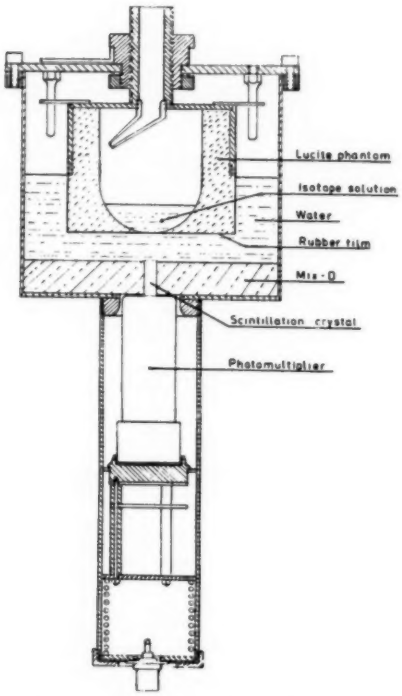


Fig. 3. The scintillation probe.

radiation); others — as in Fig. 3 — semispherical. The latter type was employed in order to get some idea of the dose distribution outside a sphere and of the degree to which it differs from that outside a flat semi-infinite source of radiation. These considerations are of medical interest because they provided a basis for estimation of the dose to the vesical mucosa on direct application of beta-emitting isotopes in the treatment of superficial multiple papillomatosis of the human urinary bladder. The measurements indicate that for spheres the dose distribution differs very little from the corresponding distribution from flat sources of radiation if the spheres have volumes of 100 cc or more; *i. e.*, volumes that are pertinent in the aforementioned treatment of papillomatosis. The deviations (for a 100 cc sphere) are measurable only in respect of $^{76}As$ (see Table 1) but are so small as to have no practical significance.

The scintillation crystal is 2 cm long and has a diameter of 0.75 cm. The photomultiplier is a Du Mont 6467 model. The multiplier current was measured with a combined voltmeter and a high tension supply as described by BELCHER (3).

Note that in Fig. 3 the isotope solution is moved, whereas in the procedure shown in Fig. 1 the position of the crystal is changed. It will readily be seen that the two modes of procedure are equivalent. In combined gamma and beta emitters the variation of the direct gamma radiation toward the photomultiplier that is caused by a maximal shift of about 0.5 cm in the isotope solution is negligible. (See Diagram 2.) The background effect may thus be regarded as constant during the experiment, provided the external environmental conditions are kept constant.

The underside of the semispherical isotope container was ground until a small hole appeared, and was then covered with a thin sheet of rubber, so stretched as to have a thickness of 10 mg/cm². The lower surface of the cylindrical container was also covered with a similar rubber sheet 10 mg/cm² thick. In order to eliminate the effect of Cerenkov radiation the crystal had to be covered with an opaque membrane. The crystal and the isotope solution were thus never closer to each other than 20 mg/cm². The absorption curve was extrapolated to zero.

## Calculation of dose

For a semi-infinite beta source the dose rate at a flat bounding surface is obtained from the formula $1{,}065 \cdot \bar{E}_\beta \cdot \sigma$ rad/hour, where $\bar{E}_\beta$ is the mean energy of the beta particles in MeV and $\sigma$ is the activity in mC per cm³ solution. The gamma dose rate on the surface of a sphere is obtained from the formula $2\pi k_\gamma \sigma a_\gamma \cdot r$, where $k_\gamma$ has been defined in Table 1; $a_\gamma$ is a factor for correction of the internal absorption, and $r$ is the radius of the sphere (16).

We have assumed that the scintillation effect is proportional to the dose (1, 2, 3, 5), and is independent of whether beta or gamma radiation impinges upon the crystal. (SOMMERMEYER 23.)

For gamma dose $D_{\gamma a}$ at a point P on the axis of symmetry $a$ cm outside a cylinder filled with isotope solution, the following calculation can be made, provided the cylinder and the surrounding medium, in the radiation sense, are equivalent to the solution itself (16, 22).

If the whole arrangement in Fig. 4 is surrounded by homogeneous material, the gamma dose in P will be

$$D_{\gamma a} = k_\gamma \sigma \int_a^l \int_0^r \frac{2\,\pi\, x\, e^{-\mu\sqrt{x^2+z^2}}}{x^2+z^2}\, dx\, dz$$

where the units of length are measured in g/cm² and $\mu$ in cm²/g; $\sigma$ is the concentration of activity in mC/ml. For $As^{76}$ $\mu = 0.07$ cm²/g, and for $Au^{198}$ $\mu = 0.105$ cm²/g. (These values are valid in respect of $H_2O$.) By substituting $\sqrt{x^2+z^2} = t$ we obtain

$$D_{\gamma a} = 2\pi k_\gamma \sigma \int_a^l dz \int_z^{\sqrt{r^2+z^2}} \frac{e^{-\mu t}}{t}\, dt$$

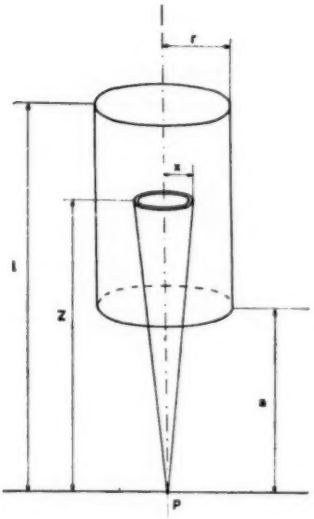


Fig. 4. Schematic representation of the principle in the derivation of the gamma-dose in a point $P$ on the symmetry axis outside a cylindrical isotope-solution.

and, if $\frac{e^{-\mu t}}{t}$ is approximated with $\frac{1}{t} - \mu + \frac{\mu^2 t}{2}$, then

$$D_{\gamma a} = \pi k_\gamma \sigma \left[ l \ln \frac{l^2 + r^2}{l^2} - a \ln \frac{a^2 + r^2}{a^2} + 2r (\tan^{-1} l/r - \tan^{-1} a/r) + \right.$$

$$\left. + \mu (l^2 - l \sqrt{r^2 + l^2} - a^2 + a \sqrt{a^2 + r^2}) - \mu r^2 \ln \frac{l + \sqrt{r^2 + l^2}}{a + \sqrt{a^2 + r^2}} + \frac{\mu^2 r^2}{2} (l - a) \right.$$

Diagram 2 shows the curve in respect of $l - a = r = 2$ cm

**Table 3**

| | $a$ cm | 0 | 0.2 | 0.5 | 1.0 | 2.0 | 3.0 | 4.0 |
|---|---|---|---|---|---|---|---|---|
| $\frac{D_{\gamma a}}{\pi \cdot k_\gamma \sigma}$ | $As^{76}$ | 4.20 | 3.02 | 2.14 | 1.35 | 0.63 | 0.36 | 0.22 |
| | $Au^{198}$ | 4.08 | 2.88 | 2.02 | 1.25 | 0.57 | 0.31 | 0.19 |

The ratio of the gamma to the beta doses at the phantom surface will be, for $As^{76}$:

$$\frac{4.20 \cdot \pi \cdot k_\gamma \cdot \sigma}{1{,}065 \cdot E_\beta \cdot \sigma} = \frac{4.20 \cdot \pi \cdot 2.0}{1{,}065 \cdot 1.17} = 0.021 \text{ and for } Au^{198} \frac{4.08 \cdot \pi \cdot 2.2}{1{,}065 \cdot 0.35} = 0.076$$

*i. e.* the gamma surface doses amount to 2 and 8 % respectively of the beta surface doses. Since the beta particles of $Au^{198}$ have a maximum range of only 0.4 cm, $l$ in Fig. 4 need not be greater than $(0.4 + a)$ cm, whereby the gamma surface dose will amount to only 4 % of the beta surface dose.

It is evident from Diagram 2 that with a shift of 0.5 cm the dose at the crystal surface facing the phantom will be reduced by 1 % of the total surface dose for $As^{76}$; whereas

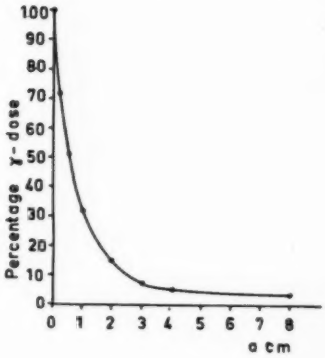


Diagram 2. The gamma dose at P in Fig. 4 at different distances from the lower surface of the cylinder as a percentage of the surface dose.

the corresponding reduction at the other crystal surface (with a crystal 2 cm long) will be only 0.08 %. From Table 3 it will be seen that the gamma depth dose curve for $Au^{198}$ diverges only very slightly from that for arsenic.

Having regard to the foregoing, it is evidently sufficient, with mixed gamma and beta emitters where the gamma contribution to the surface dose is small in comparison with the beta dose, to correct for the gamma contribution simply by subtracting the gamma dose on the basis of Diagram 2 or Table 3.

The measuring results are shown in Diagram 3.

## Film measurements

A phantom of the same type as that in the scintillation measurements was used in the film measurements. The film (Cine Positive 5302) was stacked beneath the phantom and surrounded by Mix D. Calibration was performed by exposure with the isotope solution used for exposure of the film stack, the exposure times being varied. The mean energy of the beta spectrum, as BROWNELL (7) has shown, varies fairly slowly with the absorber depth. DUDLEY (8) has demonstrated that Cine positive film is relatively energy independent for isotopes of the mean energy investigated by the present author. The energy variations of the electrons thus did not influence the calibration procedure to any substantial degree.

The films were subsequently developed in the same bath.

MARKUS and PAUL (14, 15) have shown that darkening of the film is substantially proportional to the dose in the surrounding phantom material. EHRLICH (9) has demonstrated that photographic film is energy independent in respect of gamma energies exceeding about 0.4 MeV.

Since in the present investigation it was merely a case of semi-infinite sources of radiation, *i. e.* diffuse angular distribution, no importance

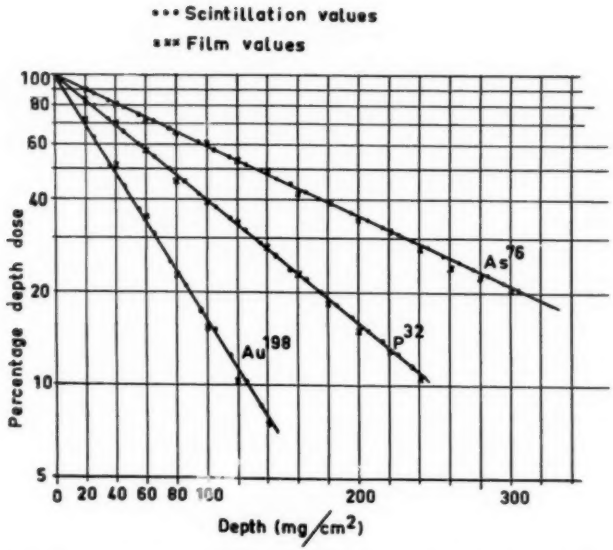


Diagram 3. Results obtained in tissue equivalent media with film and a scintillation detector.

being attached to the introduction of the non-tissue-equivalent influence of the film on the mean scattering angle, the effect of the increased back-scattering in the AgBr layer led to the choice of film having a very thin AgBr layer (0.72 mg/cm²). From formula (1) it is evident that the absorption changes rather slowly with $Z$ (since $Z/A$ is fairly constant).

A very rough estimate of the increased back-scattering through introduction of the AgBr layer was made from Diagram 1. It indicated that the increase in back-scattering was certainly less than 2 per cent of the total number of incident electrons.

The total thickness of the film was 20 mg/cm², of which the film base (triacetate) amounted to 17.8 mg/cm², and the AgBr layer together with the gelatin emulsion, 2.2 mg/cm². Darkening of the film was measured with a transmission photometer (Photovolt Corp. R 501).

The experimental results are shown in Diagram 3, where the film method is in close accordance with the scintillation method.

## Acknowledgements

I am indepted to L. G. Larsson for drawing my attention to the problem and for constant encouragement. I also wish to thank S. Benner, Mrs I. Ragnhult, and L. Jonsson for their valuable suggestions and help.

## SUMMARY

The 'tissue equivalence', in beta sense, of different chemical compounds and mixtures is discussed. A scintillation probe for beta-dose measurements is described. The depth doses from semi-infinite $As^{76}$-, $P^{32}$-, and $Au^{198}$-sources have been measured with this probe and compared with the corresponding values from film measurements.

## RÉSUMÉ

L'auteur étudie l'équivalence tissu' pour le rayonnement bêta de différents composés chimiques et mélanges. Il décrit une sonde à scintillation pour la mesure des doses bêta. Les doses en profondeur émises par des sources $As^{76}$, $P^{32}$ et $Au^{198}$ semi-infinies ont été mesurées avec cette sonde et comparées aux valeurs correspondantes données par les films dosimétriques.

## ZUSAMMENFASSUNG

Die »Gewebeekvivalenz« im Betasinne von verschiedenen chemischen Verbindungen und Mixturen wird besprochen. Eine Scintillationssonde für Messungen von Betadosen wird beschrieben. Die Tiefendosen semi-infinitiver $As^{76}$-, $P^{32}$- und $Au^{198}$-Quellen sind mit dieser Sonde gemessen und mit den entsprechenden Werten aus Filmmessungen verglichen worden.


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